Clinical Investigation Protocol for the Demonstration of Safety and Efficacy of VeSTAL Weight Loss Device in Human Subjects

Randomized Study of VeSTAL in Patients who are Overweight

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This Protocol has been read and approved by:

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1. Chief Investigator and Principal Investigator(s) Signature Page

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The undersigned have read and understood the Clinical Investigation Plan specified above and throughout pages below. In my capacity as Investigator, I understand my duties including ensuring safety of study subjects and providing study sponsor complete and timely information as outlined in this protocol. It is understood that all information pertaining to the investigational device study will be held strictly confidential. I agree to maintain the procedures required to carry out the study in accordance with accepted Good Clinical Practice principles and to abide by terms of this protocol.

Chief Investigator Signature:	

Date of Signature:	

Principal Investigator Signature (NI Site): **Date of Signature:**	

Principal Investigator Signature UCSD CCR Site: **Date of Signature:**	

Principal Investigator Signature EPARC Site: **Date of Signature:**	

Principal Investigator Signature TDE Site: **Date of Signature:**	

Abbreviations

Abbreviation	Definition
ABHI	Association of British Healthcare Industry
ACTRI	Altman Clinical and Translational Research Institute
ADE	Adverse Device Effect
AE	Adverse Event
AgRP	Agouti-Related Peptide
BMI	Body Mass Index
CA	Competent Authority
CFR	Code of Federal Regulations (United States)
CIP	Clinical Investigational Protocol
CRF	Case Report Form
CCR	Center for Clinical Research (primary trial site at UCSD campus)
CTM	Clinical Trial Mentors
C-TRIC	Clinical Translational Research and Innovation Centre
DMC	Data Monitoring Committee
DXA	Dual Energy X-Ray Absorptiometry
EPARC	Exercise and Physical Activity Resource Center (second trial site at UCSD campus)
EU	European Union
FDA	Food and Drug Administration (United States)
FFQ	Food Frequency Questionnaire
GCP	Good Clinical Practice
GVS	Galvanic Vestibular Stimulation
HISU	Human Intervention Studies Unit
IC	Informed Consent
ICF	Informed Consent Form
ICH	International Council for Harmonisation
IEC	Independent Ethics Committee
IFU	Instruction for Use (Assay/Device Handbook)
IRB	Institutional Review Board
ISO	International Organization for Standardization

IWQOL	Impact of Weight on Quality of Life
MAQ	Modifiable Activity Questionnaire
MDD	Medical Device Directive
MVe	Medial Vestibular Nucleus
NI	Northern Ireland
NPY	Neuropeptide Y
NSR	Non-Significant Risk
NTF	Note To File
PI	Principal Investigator
POMC	Pro-opiomelanocortin
REC/EC	Research Ethics Committee/Ethics Committee
SADE	Serious Adverse Device Effect
SAE	Serious Adverse Event
SAP	Statistical Analysis Plan
SOP	Standard Operating Procedure
SR	Significant Risk
TDE	Texas Endocrinology and Dibates
tES	Transcranial Electrical Stimulation
TBWL	Total Body Weight Loss
TLS	Transport Layer Security
TSP	Test Site Protocol
UADE	Unanticipated Adverse Device Effect
UCSD	University of California San Diego (refers to primary site CCR and EPARC)
UK	United Kingdom
USA	United States of America
UU	University of Ulster
VeNS	Vestibular Nerve Stimulation
WST	We Slim Together

2. Statement of Conformity

Neurovalens's representatives declare that the device specified for the Clinical Study Investigation Plan for Investigational Analysis of patients using the VeSTAL device for weight loss was designed to conform with the essential requirements of the Medical Device Directive 92/43EEC (MDD) with the exception of those requirements which the Clinical Study is intended to fulfil and that every reasonable precaution has been taken to protect the health and safety of the subjects.

3. Study Synopsis

TITLE	A randomized, double blind sham controlled clinical trial to evaluate the efficacy of vestibular nerve stimulation (VeNS), combined with a lifestyle modification program, compared to a sham control and a lifestyle modification program as a means of reducing excess body weight and body fat.
FACILITIES	All study visits will be carried out between four sites: 1. Altman Clinical and Translational Research Institute's Center for Clinical Research, which is part of the La Jolla campus of University of California San Diego (CCR) 2. Exercise and Physical Activity Resource Center (EPARC), CALIT2 Building, Rooms 3401-3506 which is also part of La Jolla campus of UCSD. 3. Human Intervention Studies Unit (HISU) at University of Ulster (UU) Coleraine Campus in Northern Ireland 4. Texas Diabetes & Endocrinology site in Austin Texas (TDE) Trial sites CCR and EPARC at the UCSD campus will be referred to collectively as 'UCSD' throughout this protocol. An unfunded collaboration agreement will be established between all sites.

	There is a DXA scanner located at all sites described above, apart from the TDE site where an external contractor will be used (see Section 6.5).
NUMBER OF SITES	4
STUDY DESIGN	The aim of this study is to evaluate the efficacy of non-invasive electrical vestibular nerve stimulation (VeNS), together with a lifestyle modification program, as a method of reducing excess body weight and body fat, as compared to a sham control with both study arms incorporating a lifestyle modification program. • Allocation: Randomized • Endpoint classification: Efficacy Study • Intervention Model: Parallel Assignment in 1:1 active to control allocation • The aim of the study is to recruit a total (i.e. across all 4 sites) of 200 participants that pass the screening process and are randomized into the treatment protocols. With a dropout allowance of 10% this should generate a minimum of 90 active treatment and 90 control subjects. • Masking: Double Blind (Subject, Nursing staff, Dietician, Co-coordinators, Outcomes Assessor and any other study staff who have contact with the subject) • Data from all sites will be collated at the end of the studies and analysis will be performed on one data set. This protocol governs the activities at both the USA and NI/UK clinical sites.
PRIMARY OBJECTIVE	To establish the clinical performance of the VeSTAL device stimulation effect in patients who are overweight.

SECONDARY OBJECTIVES	To evaluate between baseline and 6 months the safety of the VeSTAL device relative to control group, in terms of the occurrence of adverse events, changes in vital signs (blood pressure and heart rate), and hearing tests. A variety of secondary endpoints will also be assessed statistically at the 6-month timepoint after the primary endpoints. The following of these may form the basis of potential future labelling claims: • Total body fat loss (as measured by whole body DXA scanning); • Healthy Eating Index (HEI) score (https://epi.grants.cancer.gov/hei/) (as measured by the two-day 24-hour dietary recall); • Total caloric intake (as measured by the two-day 24-hour dietary recall); • Systemic inflammation (as measured by high-sensitivity C-reactive protein); • Atherogenic index (as determined by the ratio of total cholesterol to HDL); • Quality of life (as measured by the IWQoL questionnaire). The full details of the testing procedures procedure for these endpoints will be laid out in the Statistical Analysis Plan. All other secondary endpoints at the 6-month timepoint will be hypothesis generating only.
NUMBER OF SUBJECTS	In order to achieve adequate numbers, we estimate that we may have to consent (i.e. enrol into the study) up to 120 subjects at the CCR site, approximately 80 subjects at the EPARC, up to 40 patients at the TDE site, and up to 150 subjects at the UU site. So a total of about 300 across all sites. This is because the baseline screening can only take place after the formal consent process. The anticipated recruitment across the study sites will be as follows: o CCR: up to a total of 106 subjects who pass through screening and are randomized;

	○ EPARC: up to a total of 60 subjects who pass through screening and are randomized; ○ UU: up to a total of 94 total subjects who pass through screening and are randomized. ○ TDE: up to a total of 40 total subjects who pass through screening and are randomized.
SUBJECT SELECTION CRITERIA	**Pre-screening of Subjects** In order to prevent subjects traveling in needlessly to the study site it is proposed that the study staff allow prospective subjects to pre-screen themselves over the phone. This pre-screening will involve the study coordinator reading out the inclusion and exclusion criteria to prospective subjects during an initial phone call. The screening questionnaire will not be completed at this stage. Rather the purpose is to make prospective subjects aware as to what the inclusion and exclusion criteria are, since there is no point in them wasting their time in coming in if they are ineligible. Moreover, this will also allow study coordinators to provide prospective subjects with a verbal description of the study to the subjects. For the UU site – this pre-screening activity will be managed by Compliance Solutions Life Sciences Ltd. and the UU study team with the names of potentially eligible subjects being passed directly to the site. Potentially eligible participants who successfully meet the Pre-Screening inclusion criteria will be sent a study information pack. This study information pack will include the following documents: Informed Consent Form, Food Frequency Questionnaire, Modifiable Activity Questionnaire, Two-day 24 Hour Recall, Concomitant Medications Form and Inclusion/Exclusion Form. The participant will be asked to bring these completed forms with them to their baseline visit (Visit 1, 0 Months) and at this visit each form will be reviewed by the study staff.

Inclusion/ Exclusion Criteria Questionnaire

Subjects who have signed the Informed Consent Form (ICF) will be asked to complete a Screening Questionnaire to ensure that they meet to requisite inclusion criteria. Consideration was given to which of the exclusion criteria for which it would be appropriate, and practical, to actively screen. Consideration was given to harm mitigation as frequently obese subjects have mild irregularities of blood tests (e.g. liver function) that are no, or limited, clinical relevance, and clearly it would not be desirable to unnecessarily exclude subjects, nor cause them undue anxiety, on this basis. As detailed below, it was decided that the appropriate conditions to actively screen for are diabetes mellitus, hypothyroidism and pregnancy.

Pre-enrollment Screening for Type 2 Diabetes Mellitus

After being consented but prior to being randomized to a treatment arm, potential subjects will be screened for type 2 diabetes mellitus by means of a fasting blood glucose test. This will be done via a finger prick sample and glucose meter. Testing should be performed in the morning after an overnight fast of at least 8 hours.

If a subject is found to possibly have type 2 diabetes [i.e. they have a reading of 126 mg/dl (7 mmol/L) or above] during the initial screening, then they will be excluded and asked to notify their primary care physician. However, if a subject passes the initial screening but then a suspicion of this diagnosis is made during one of the study tests, then the subject will be asked to notify their primary care physician, though they will still be allowed to complete the study, and a note to file will be made.

Screening for Pregnancy

Female subjects of childbearing potential (i.e. who have not gone through the menopause or had a hysterectomy or oophorectomy) will be screened for pregnancy by means of a standard urinary pregnancy test. It is important to

	know in a trial of an investigational device for weight loss if a female subject has become pregnant, and especially so at 0 and 6 months when the subjects undergo a DXA scan which involves a small dose of ionizing radiation. Thus urinary pregnancy tests will be carried out at 0, 3 and 6 months. Female subjects will be requested, as part of the consent process, to refrain from becoming pregnant for the duration of their participation in the study. Should a subject become pregnant after enrollment then they will be withdrawn from the study, both because of the use of ionizing radiation in DXA scans and the increase in weight that occurs during pregnancy. **Screening for Hypothyroidism** After being consented but prior to being randomized to a treatment arm, potential subjects will be screened at the baseline visit for hypothyroidism. This will be done using a finger prick test for hypothyroidism, which is available for clinical use in the USA and NI sites as ThyroChek (See: http://thyrochek.com). This product has a sensitivity of 98.5% and a specificity of 96.9%. It is a qualitative test that takes about 10 minutes to give a result. If positive it indicates that the TSH level is > 5 mIU/L. If a subject tests positive, then they will be excluded from the study and asked to see their primary care physician to be investigated for hypothyroidism. **Subject Eligibility** Where a subject fails to fulfill the inclusion/ exclusion/ screening criteria, this will be documented, and the investigator will retain the signed consent form. The subject will not advance any further into this clinical investigation.
TEST PRODUCT, HOW USED	Study staff will train the subject on how to use their study device. Specifically, this will include: how to first prepare the skin over the mastoid processes with an alcohol wipe; a demonstration of where to place the 32mm diameter hydrogel electrodes (i.e. behind the ears on the skin overlying the mastoid processes – see Figure 1); how to access the VeSTAL study app and pair the iPod (using Bluetooth Low Energy) to the device; how to operate the device

	using the app; and how to use the up/down buttons on the device if unable to use the study app. Study staff will provide subjects with a printed copy of the Instructions for Use (IFU) and also highlight its location as a PDF on the iPod. The Device Use Schematic will also be highlighted to the subjects – it will only be as a PDF on the iPod. Also study staff will demonstrate to the subjects: how to charge the device; to dispose of used electrodes after a stimulation session; how to store unused electrodes in a sealed manner so they do not dry out; how to turn the device on and off using the power button; and how to use the app or power button to pause a stimulation session. Subjects will be asked to try and use their allocated device for an hour every day, and at least five times a week. Theoretically it is thought that the device may be more effective if used while sitting upright (as opposed to lying flat) (Macefield & James, 2016), due to the orientation of the otolith organs. As such subjects will be encouraged to use the device while sitting upright, and also in the evening as sometimes people can feel a bit soporific after vestibular stimulation. Subjects will be instructed not to walk around, operate machinery or drive while using their device. Most people can tolerate up to 1.0mA without difficulty and subjects will be encouraged to turn the device up as much as they are comfortable with.
DURATION OF SUBJECT PARTICIPATION AND DURATION OF STUDY	Subjects will be enrolled at a rate of up to 3 subjects per day from the study start date. The study will last approximately 6 months. The final analysis will be conducted at the 6 month timepoint, and this will form the basis of a future de novo application. Participants will be given the headset device to use in the home environment for a period of 6 months, the daily use of the device will be recorded automatically via the device application. Both the UCSD CCR site (based in

	USA) and the UU site (based in NI) started in September 2019. The EPARC site is expected to start in early 2020.The TDE site (based in the USA) is expected to start after September 2020.
OTHER EVALUATIONS	VeSTAL design has been developed in compliance with the design control requirements contained in 21 CFR Part 820.30 and ISO 13485:2016.
STATISTICS **Primary Analysis Plan Details**	Two statistical analysis pathways have been identified each separate for the EU and USA regulatory clearance purposes.
STATISTICS **Primary Analysis Plan for Regulatory Clearance pathway in USA**	The Primary Analysis population will be 200 subjects, from the data collected across two sites details as given above. **Primary Endpoints:** The efficacy of the VeSTAL device within the treatment group will be quantified by calculation of two primary endpoints after 6 months. 1. mean percent weight loss; 2. proportion of participants who lose at least 5% body weight. **Study Efficacy Acceptance Criteria:** • Mean: There should be at least a 2% total body weight loss (TBWL) superiority margin between the active-product and sham-treated groups. • Categorical: The proportion of participants who lose 5% TBWL or more in the active-product group is at least 50%, independent of the sham control Both weight loss efficacy acceptance criteria must be met to claim success. These criteria can be specified in mathematical formulae as follows: Rejection of both the null hypotheses $\mu A - \mu C \leq 2\%$ and $\pi A \leq 50\%$ at the 2.5% (1-sided) level Where:

	μA and μC are the underlying population mean percentage weight loss on active and control respectively πA is the underlying population proportion of subjects who lose ≥5% of baseline body weight on active. **Methods: Primary Aims** For total body weight loss (TBWL) as a continuous measure, we will use a linear regression with TBWL as the dependent variable and group and other covariates of interest (gender, age, etc.) as independent variables. To assess any longitudinal effects, we will use a linear mixed effects model for weight over time, with fixed covariates for baseline weight, time, group, and a time / group interaction, and a random intercept to account for within-subject correlation. We will use these models to generate two-sided 95% confidence intervals for the difference in mean percent weight loss. We will use a one-sided exact binomial test to determine if the response rate in the treatment group significantly exceeds 50%, with response defined by a loss of at least 5% of their body weight. We will analyze mean fat loss in the same way as TBWL as a continuous measure with linear regression and linear mixed effects models. Analyses will be conducted at 6 months.
STATISTICS **Primary Analysis Plan for Regulatory Clearance pathway in EU**	The Primary Analysis population will be 200 patients, from the data collected across two sites details as given above. Primary Endpoint: The efficacy of the VeSTAL device within the treatment group will be quantified based on a single primary endpoint: 1. Mean percent weight loss

	Study Efficacy Acceptance Criterion: • Mean: There should be at least a 2% total body weight loss (TBWL) superiority margin between the active-product and sham-treated groups. This criterion can be specified in mathematical formulae as follows: mA – mC ≥ 2% and pt ≤ 0.05 Where: mA and mC are the observed mean percentage weight loss on active and control respectively pt is the p-value (two-sided) from a linear regression comparing percentage weight loss between active and control (H0: μA = μC; H1: μA ≠ μC) μA and μC are the underlying population mean percentage weight loss on active and control respectively This single study efficacy acceptance criterion for the EU regulatory clearance pathway will be treated to the same statistical analysis as the mean criterion for the USA regulatory clearance pathway.
Rationale for Number of Subjects	The primary outcomes are TBWL from the start of treatment (as a percentage of baseline total body weight), and the proportion of subjects who lose at least 5% of their baseline total body weight. These outcomes will be assessed at 6-months. Power calculations are based on simulation with assumptions informed by pilot data and the Modius weight-loss device as follows: • 90kg median baseline weight in both groups (log-normal distribution with CV=24%) • 6.6% mean percent weight loss in the active group at 6 months (SD 3.9%),

	• 3.1% mean percent weight loss in the control group at 6 months (SD 3.9%), • correlation between baseline weight and weight loss at 6 months of -0.4. Under these assumptions, with a sample size of 180 participants randomized 1:1 active: placebo, there is greater than 80% power to demonstrate both the weight loss mean and categorical acceptance criteria at the 6 month timepoint.

4. Purpose and Scope

This protocol describes the overall device investigation; governs the activities at the site of the device investigation and describes the study details, interactions and records to be generated throughout the study. This document serves as the lead document and supersedes all other documents. e.g. Instructions for Use (IFU).

There is an ongoing and worsening problem with obesity in the developed, and much of the developing world. Although it has long been realized that Western diets that are rich in sugar and fat play an important role in this, it has only recently been realized that exposure to these diets, particularly in childhood, can damage the part of the brain that determines how much fat there is in the body. The result of this damage is that the so-called “set-point” for fat in this part of the brain is pushed upwards. There is a lot of evidence from animal studies that activating the brain’s balance (vestibular) system pushes this set-point for fat downwards to cause fat loss, probably because this tricks the brain into thinking that the animal is more physically active. The aim of this study is to determine whether the same effect can be triggered in humans by non-invasively stimulating the vestibular system with a small electrical current through the skin behind their ears, in order to evaluate the efficacy of non-invasive electrical vestibular nerve stimulation (VeNS), together with a lifestyle modification program, as a method of reducing excess body weight and fat, as compared to a sham control and lifestyle modification program.

This protocol will be applied at the following sites:

1. Ulster University, Cromore Road, Coleraine, County Londonderry, BT52 1SA in Northern Ireland, at the Human Intervention Study Unit (HISU)
2. Altman Clinical and Translational Research Institute (CCR) building which is part of the La Jolla campus of UCSD
3. Exercise and Physical Activity Resource Center (EPARC), CALIT2 Building, Rooms 3401-3506 which is also part of La Jolla campus of UCSD.
4. Texas Diabetes & Endocrinology 5000 Davis Lane, Suite 200 Austin, TX 78749 USA

All sites will be randomized at 1:1 (active: sham) and an endpoint analysis will be performed at 6 months.

5. Background

The VeSTAL device utilizes a technology called galvanic vestibular stimulation (GVS) (sometimes termed vestibular nerve stimulation (VeNS)). In the envisaged configuration, the device will be placed on the head in a manner analogous to headphones and will deliver a small electrical current (1.0mA or less) to the skin behind the ears, over the mastoid processes.

The VeSTAL device can only be used for up to one hour a day, and after being used for this time period, it will automatically lock out until the next day. Delivery of electrical current to the skin over the mastoid region is known to activate all five components of the vestibular apparatus, but lower level currents (below 3mA) are thought to particularly activate the two otolith organs responsible for detecting linear acceleration and gravity (Zink, Bucher et al., 1998). It is these otolith organs that, from the animal studies, are particularly associated with a reduction in body fat. This technique – known as bilateral bipolar galvanic vestibular stimulation – has been known since the 19th century and is thought to be safe (Fitzpatrick and Day, 2004).

5.1 *Published Clinical Evaluations to Support Vestibular Stimulation and Weight Loss*

There is growing evidence for a "set-point" in the hypothalamus that acts to regulate body mass composition, including body fat (Harris, 1990) (Horvath, 2005) (Yon, Mauger et al., 2013). This feedback control mechanism modifies feeding behavior and metabolic rate, in order to maintain body mass composition within predetermined parameters and does so in order to optimize energy homeostasis. As such, deviations too far in either direction

from the set-point are strenuously resisted (Sims, Horton et al., 1968) (Horvath, 2005), (Yon, Mauger et al., 2013). Thus, not only is it hard to change body mass composition via diet and exercise, but even if one can, maintaining the new leaner composition in the long term is typically doomed to failure, as the brain, in effect, fights against the change.

Several nuclei within the hypothalamus are involved in regulating this set-point, but one particularly vital one appears to be the arcuate nucleus with its populations of pro-opiomelanocortin (POMC), and agouti-related peptide (AgRP) / neuropeptide Y (NPY) co-expressing neurons (Parton, Ye et al., 2007, Belgardt, Okamura et al., 2009, Varela and Horvath, 2012) (Wang, He et al., 2015). These neurons together comprise the central melanocortin system, which acts to regulate energy homeostasis, by responding to circulating hormones, nutrients, and neuronal inputs (Cone, 2005). In the case of the POMC neurons, the response is anorexigenic with a decrease in food intake and an increase in energy expenditure. Conversely, the orexigenic AgRP/ NPY neurons act in an antagonistic manner (Cone, 2005) .

The set-point for body mass composition that is determined by the central melanocortin system obviously varies from person to person, and is influenced by both genetic and epigenetic factors (Herrera, Keildson et al., 2011). However, it appears that exposure to Western diets, particularly during childhood and adolescence, with their often excessive quantities of sugar and saturated fatty acids, can damage neuronal populations within the hypothalamus and push the set-point for body fat upwards (Horvath, 2005) (Parton, Ye et al., 2007, Yon, Mauger et al., 2013). This makes the overweight body composition become the normal body composition as far as the brain is concerned, and reverting to a leaner set-point once this has occurred can be difficult (Horvath, 2005). The set-point is not totally immutable though, as there is evidence that it can be lowered by long term dietary changes, such as, consuming a lot of polyunsaturated fatty acids (Yon, Mauger et al., 2013). Similarly, it is also now thought that bariatric surgery acts by reducing the set-point (presumably via hormone release), rather than simply decreasing caloric intake (Hao, Mumphrey et al., 2016).

The clear ability of vestibular stimulation to modify body mass composition (most notably to reduce body fat, but also to increase lean muscle mass and bone mineral density), appears to be mediated via an otolith-vestibulo-hypothalamic pathway (Fuller, Jones et al., 2004) (Fuller, Warden et al., 2000). This reflects the vestibular system's vital role in homeostasis and, indeed, vestibular input is known to project to multiple brainstem homeostatic sites (Balaban and Yates, 2004) (McGeoch, 2010). This includes the arcuate nucleus of the hypothalamus (Fuller, Jones et al., 2004), which, as stated, sits at the crux of the central melanocortin system. Moreover, three further observations confirm an intimate interplay between the vestibular and central

melanocortin systems: First, melanocortinergic neurons have been found in the medial vestibular nucleus (MVe) (Shang, Xiong et al., 2015); second, an enzyme important in the modulation of melanocortin signaling is expressed in vestibular nuclei, in particular MVe (Jeong and Diano, 2014); and third, POMC neurons in the nucleus of the solitary tract, which are part of the central melanocortin system that connects to the parasympathetic nervous system, also receive input from the MVe (Wang, He et al., 2015).

The prominent role of the MVe here, is noteworthy, as it is the main projection target of the utricle (Newlands, Vrabec et al., 2003), which is the otolith organ that detects movement on a horizontal plane. Also, retrograde tracer studies have identified the MVe as one of the areas involved in the central control of the sympathetic nerves supplying both normal fat (properly termed white adipose tissue) (Bamshad, Aoki et al., 1998), and brown adipose tissue (Bamshad, Song et al., 1999), which is a more specialized type of fat extremely rich in mitochondria, the organelles of the cell that generate energy. It is believed that activating brown adipose tissue could act to reduce the total amount of white adipose tissue, and thus have an anti-obesity potential, and moreover, insulin sensitivity and therefore type 2 diabetes mellitus (Scheele and Nielsen, 2017). There are also definite but, as yet, somewhat anatomically less well delineated connections between the vestibular nuclei in the brainstem and the sympathetic nerves that innervate skeletal muscle and bone (Vignaux, Besnard et al., 2013) (Bolton, Wardman et al., 2004, Bent, Bolton et al., 2006)

In summary, it thus seems that the central melanocortin system interprets activation of the MVe (by horizontal movements stimulating the utricle) to indicate a state of increased physical activity. Homeostatically, in such a state of apparent increased activity, it is optimal, from an energy conservation point of view, for the body to have a leaner composition, in order to reduce unnecessary energy expenditure from carrying around excess fat. Also, under such conditions it would be homeostatically more efficient for the muscles and bones to be stronger, as this would facilitate them coping with the state of chronically increased movement. Given its described connections to both sides of the autonomic nervous system (i.e. anorexigenic POMC neurons within the parasympathetic nervous system (Wang, He et al., 2015), and the sympathetic nerves innervating white and brown adipose tissue (Bamshad, Aoki et al., 1998) (Bamshad, Song et al., 1999), bone and muscle (Bolton, Wardman et al., 2004, Bent, Bolton et al., 2006) (Vignaux, Besnard et al., 2013), the MVe is also well placed to effect these changes in body mass composition – i.e. reduce total body fat and increase lean muscle mass and bone mineral density.

Thus GVS, which is known to stimulate the otolith organs (Zink, Steddin et al., 1997) (Fitzpatrick and Day, 2004), when repeatedly administered over time, will cause recurrent activation of the MVe, which will be taken

by the central melanocortin system to indicate a state of chronically increased physical activity, with the consequence of altering the set-point for body mass composition to a physically leaner state; with less adipose tissue and stronger bones and muscles.

Pilot data collected at UC San Diego (IRB study #110538) demonstrated a reduction in body fat when receiving vestibular stimulation over a 4 month period. Total body and truncal fat was measured using DXA scanning. The treatment of 3 males and 3 female subjects of median 23 years old, showed a significant reduction in truncal fat ($t(7) = 2.88$, $p < 0.025$), the data also suggested a trend towards a significant reduction in total body fat ($t(7) = 2.17$, $p = 0.067$). Overall the treatment group showed an 8.3% decrease in truncal fat (cf. an 8.6% increase in the control group), and a reduction in total body fat of 6.3% (cf. a 6% increase in the controls). The control group participants received a sham device stimulation, the overall comparison was skewed due to a subject who gained 3.8kg in truncal fat. However, if this subject is considered as zero weight gain the difference in truncal fat between groups remains significant ($t(7) = 2.53$, $p = 0.039$) (McGeoch, McKeown et al., 2017)

5.2 *Studies on Galvanic Vestibular Stimulation*

Krystal et al. (2010) investigated the effect of one hour of GVS, at up to 0.5mA delivered at 0.5Hz, in 101 healthy volunteers and 97 controls who received sham stimulation. They found that the stimulation was “generally well tolerated.” Adverse events occurred in 16.8% of the treatment group compared to 6.2% of sham treated subjects. Headache was the commonest side-effect which occurred in 11.9%, with dizziness the second most common at 3%. In total 12 of the 17 adverse events were ranked as mild, 4 moderate and 1 episode of dizziness and nausea as severe. All of the adverse events resolved spontaneously or, in the case of headache, with the use of over the counter medications. The investigators note that “no serious or unanticipated device related effects” occurred. It is noteworthy that the total current delivered by the GVS device in this study is directly comparable to that delivered by the VeSTAL device, as it has a 50% duty cycle, which means it delivers current only half the time (Krystal, Wyatt et al., 2010).

Marshall et al. (2010) described the effect of GVS in a population of 105 individuals with chronic insomnia. The device used in their study was similar to the VeSTAL device in that it could deliver between 0.1 and 1.0mA at 0.5Hz. However, unlike the VeSTAL device it did not have the 50% duty. The study participants all received GVS for an hour a day for 30 days. Only two adverse events (a petit mal seizure and tinnitus) were reported in the group, neither of which was thought by the investigators to be attributable to use of the GVS device. (Marshall, Jasko et al., 2010)

A study in which noisy GVS was delivered to 30 elderly people at between 0.1 and 0.2mA for a continuous three-hour session also observed no adverse events (Fujimoto, Yamamoto et al., 2016). The word noisy here indicates that the stimulation current is varied randomly. Although this is clearly a different waveform from that delivered by VeSTAL, the total current delivered over the three-hour period will, because of the 50% duty cycle in VeSTAL's waveform, be similar.

There is thus a body of literature in the adult population supporting the safe usage of both transcranial electrical stimulation (tES) and more specifically GVS devices delivering at least 1mA in stimulation. In conclusion, there is not a significant risk of harm from using the device, and as such criteria in the FDA guidance document do not apply. Thus, as the device does not meet the criteria of a significant risk (SR) device it is a non-significant risk (NSR) device.

5.3 *Risk Evaluation of Published Literature Vestibular Stimulation*

The following risk assessment was also carried out for the vestibular nerve stimulation device, and its embedded software, that can deliver up to 1mA. Under the guidance for IRBs from the FDA a device will be determined to be non-significant risk (NSR) if it does not meet the definition of a significant risk (SR) device (see https://irb.ucsd.edu/device.pdf). A SR device is one which meets at least one of the following criteria:

a. The device is intended as an implant.
b. The device supports or sustains human life.
c. The use of the device is of substantial importance in diagnosing, curing, mitigating, or treating disease or preventing impairment of health.
d. The device could cause significant harm to any subjects.
e. The subject must undergo a procedure as part of the device study.
f. The device appears on the FDA list of significant risk devices.
g. The study or any of the study procedures could cause harm to the subjects which:
    i. could be life threatening;
    ii. could cause permanent impairment of a body function;
    iii. could cause permanent damage to body structure;
    iv. or could necessitate medical or surgical intervention to preclude permanent impairment of a body function or preclude permanent damage to body structure.

As the device is not implanted, no procedure is required to use it, it does not support or sustain life and it is not on the FDA list of significant risk devices, criteria a, b, e and f clearly do not apply. Regarding criterion c, the aim of this study is to investigate whether vestibular stimulation can, in conjunction with lifestyle interventions, assist with weight and fat loss. It may be the case that VeNS has such a role, but it is unlikely that this will be of "substantial importance in … curing, mitigating, or treating disease, or preventing impairment of health." Thus, criterion c does not apply.

This then leaves criteria d and g, which both pertain to users suffering significant harm from the device. When the device is turned on it will start at 0mA and the current can then be increased in 0.1mA increments up to 1.0mA. The waveform is a bipolar rectangular shape with 50% duty cycle and – i.e. half the time no current is being delivered. The device is powered via a 3.75V battery and can be recharged through a USB connection. There is also a hardware interlock to ensure the device cannot be switched on when the charging cable is being used. Note the device is battery, and not mains, powered.

The devices are placed on the head in a manner analogous to headphones, and the VeNS devices will deliver a small electrical current (1.0mA or less) to the skin behind the ears over the mastoid processes. The device can only be used for up to one hour a day, and after being used for this time period the device will automatically lock out for the next 16 hours. Delivery of electrical current to the skin over the mastoid region is known to activate all five components of the vestibular apparatus, but lower level currents (below 3mA) are thought to particularly activate the two otolith organs responsible for detecting linear acceleration and gravity (Zink et al., 1998). It is these otolith organs that, from the animal studies, are particularly associated with a reduction in body fat. This technique – known as bilateral bipolar vestibular stimulation – has been known about since the 19th century and is known to be safe, though skin irritation behind the ears can occasionally occur (Fitzpatrick & Day, 2004).

In 2015, Paneri et al. (2015) reviewed the safety of repeated sessions of transcranial electrical stimulation (tES), a term that they use to encompass both the transdermal electrical modulation of cranial nerves (i.e. as in VeNS) and transcranial direct current stimulation (tDCS) of the cerebral cortex. Based on studies of tES on patient groups with depression (Brunoni, Valiengo et al., 2013) and migraine (Magis, Sava et al., 2013), and an array of studies of tES on normal volunteers (Brunoni, Amadera et al., 2011) (Kessler, Turkeltaub et al., 2012) (McIntire, McKinley et al., 2014) (Morales-Quezada, Cosmo et al., 2015) (Nitsche, Liebetanz et al., 2003) (Poreisz, Boros et al., 2007) (Raimundo, Uribe et al., 2012) (Russo, Wallace et al., 2013) (Tadini, El-Nazer et

al., 2011), together with theoretical assessments of tES (Bikson, Datta et al., 2009) (Brunoni, Nitsche et al., 2012), they commented that the “safety and tolerability profile previously accumulated regarding extended use of tES in clinical populations is compelling and supports a low-risk or non-significant risk designation” (Paneri, Khadka et al., 2015)

Paneri et al. (2015) also assessed the safety and tolerability of repeated tES across time in a group of 100 volunteers who were split into three groups. The first group received sham stimulation, the second tES at 2mA, and the third pulses of tES at between 5 to 7 mA (this is notably higher than the 1mA maximum we are proposing to use). A total of 1905 treatment sessions were carried out in total across the three groups. The investigators report no serious adverse events in any treatment condition, and that the common side effects were restricted to skin tingling, itching, and mild burning. Moreover, the incidence of these events were not statistically higher in the tES groups (Paneri, Khadka et al., 2015). These mild skin sensations were not associated with withdrawal from the study and the Investigators reported they became less salient after the first few sessions. Other adverse events, such as headache, were rare (<5%) and statistically indistinguishable across the three groups. Paneri et al (2015) concluded that the “repeated use of limited output tES across extended periods, is well tolerated and poses no significant risks to healthy subjects, as previously observed in clinical studies”.

This conclusion is in keeping with the findings of Wilkinson et al. (2009), who reported on a stroke patient who received repeated VeNS sessions as part of his rehabilitation therapy. The Investigators observed no adverse events during stimulation over 5 consecutive daily sessions of VeNS at 1mA for 30 minutes per day. Utz et al. (2011) studied the adverse effects of 255 VeNS sessions at 1.5mA (again higher than the 1mA we propose to use) in 55 stroke patients and 30 healthy controls. They found only a few mild adverse effects, with the most common being slight itching (mean 10.2%) and tingling (mean 10.7%) underneath the electrodes. They concluded that VeNS induces “very few and mild adverse effects in healthy and persons with stroke and [is] safe” (Utz, Korluss et al., 2011). There is thus a body of literature supporting the safe usage of both tES and specifically VeNS devices delivering at least 1mA in stimulation.

Similarly, in February 2016, Halo Neuroscience, a company based in San Francisco, released additional data on the safety of tES; on this occasion for a 2mA tDCS device used on 1010 subjects (HaloNeuroscience, 2016) They state that “there were zero reports of burn or seizure activity across all 1010 subjects, zero serious or unexpected adverse effects and zero withdrawals due to adverse events other than unpleasant sensation”.

They conclude that their tES study showed “a favorable safety profile and [was] associated with a very low incidence of adverse events, with no unexpected or serious adverse events”.

The above studies were carried in the adult population, However, there is also a smaller body of work reporting the use of VeNS in the under 18 age group. Two studies have been published looking at the effect of VeNS (GVS) on adolescent scoliosis (Pialasse, Descarreaux et al., 2015) (Pialasse, Descarreaux et al., 2015). These studies both administered VeNS up to 1mA in intensity in several dozen children aged from 10 to 18 years. Both children with adolescent scoliosis and healthy controls were recruited. Binaural, bipolar stimulation was administered up to 1mA in repeated pulses of 2 seconds in duration. No adverse events were reported by the investigators. Similarly, another study involving both children and adults, undergoing vestibular rehabilitation, who were repeatedly administered one-minute of VenS up to 2mA in intensity, also did not document any adverse events (Carmona, Ferrero et al., 2011).

Specific measures taken by the study device to reduce the incidence of skin irritation are the 50% duty cycle (i.e. half the time no current at all is being delivered), and the maximum current of 1.0mA. This means that the maximum phase current that can be delivered is only 500μC. It is noteworthy that no duty cycle, with the consequent reduction in maximum phase current, was reported as being used in any of the studies describing the use of VeNS in the pediatric age group (Carmona, Ferrero et al., 2011) (Pialasse, Descarreaux et al., 2015, Pialasse, Descarreaux et al., 2015), and despite, this none of them reported any adverse effects. In conclusion, there is not a significant risk of harm from using the device, and as such criteria d and g do not apply. Thus, as the device does not meet the criteria of a significant risk (SR) device it is a non-significant risk (NSR) device.

6. Overview of Clinical Study

Note: This protocol refers to ‘study staff’ throughout. The study staff role may be filled by clinical trial coordinators, or other clinical trial site staff who may be trained technicians, nurses or dieticians qualified to perform the various study activities detailed throughout this protocol. Each subject will be assigned a unique subject identifier (Subject ID). Individual subjects are allowed to participate in the study only once.

The purpose of this investigational device study is to collect data to support regulatory submissions, primarily in the United States of America (USA), but it may also be used to support submissions in other regions, including the European Union (EU).

This double blind randomized controlled trial will be conducted at four clinical trial sites: The University of California, San Diego's Clinical (UCSD) and Ulster University (UU) in Northern Ireland. The study is an Investigator Initiated Study and when the term 'Study Sponsor' is referenced, it relates to Neurovalens Ltd. (the device manufacturer). The study site located in USA will enroll approximately 106 subjects post-randomization. The study site in Northern Ireland will enroll approximately 94 subjects post-randomization. Each Subject will be assigned a unique Subject identifier (Subject ID). Individual subjects are allowed to participate in the study only once. The study will be randomized for a 6 month period with a 1:1 active to sham device allocation. After completion of this initial 6 month period, the study will stop.

Screening of subjects will be completed remotely over the telephone with the completion of a study inclusion and exclusion criteria Screening Questionnaire and willingness to participate in the study. This interview will be conducted via the clinical sites telephone line which will be detailed on the advertisements associated with advertising the study for identification of potential study participants. Subjects will be given a brief outline of the study and asked if they would be interested to continue for potential inclusion by answering inclusion and exclusion questions, all interviews will be recorded on the study Pre-Screening questionnaire (telephone screening form). Subjects who are deemed eligible on paper will be asked for their contact details and this information will be forwarded to the clinical site to arrange the 1st baseline visit activities (Visit 1, 0 months).

At the UU site only: If a particiant has successfully met the inclusion criteria via telephone Pre-Screening, their contact information will be sent via email to the study staff at UU to arrange for their baseline visit (Visit 1, 0-Month). The participant will be informed that a study information pack will be sent to them for completion prior to their baseline visit. This study information pack will include the following documents: Informed Consent Form, Food Frequency Questionnaire, Modifiable Activity Questionnaire, Two-day 24 Hour Recall, Concomitant Medications Form and Inclusion/Exclusion Form. The participant will be asked to bring these completed forms with them to their baseline visit (Visit 1, 0 Months) and at this visit each form will be reviewed by the study staff at UU.

The scheduled site visits that each subject will undertake are:

- Site Visit 1 (~3.5 hours; at 0 month timepoint for enrollment) (Subject fasted for at least 8 hours)
- Site Visit 2 (~1.5 hours; at 3 month timepoint) (No fast required)
- Site Visit 3 (final visit: ~3 hours; at 6 month timepoint) (Subject fasted for at least 8 hours)

The subject will be asked to refrain from exercise during the 24 hours period before each of the visits that involve DXA scanning (i.e. Visits 1 and 3).

There is also a scheduled contact (email or telephone call) with the subject at the 1 month timepoint to ask subjects to complete the Adverse Event questionnaire and Blinding assessment (gathered by the VeSTAL study app).

At the end of Visit 3 (i.e. only after the above actions have been completed), the final analyses will be conducted. Subjects will return their devices and cease participation at Visit 3.

6.1 *Summary of Site Visit 1 Screening / Baseline (0 months)*

- The study staff will provide potential subjects with a description of the study, including requirements for participation, specific study activities, and procedures, these procedures should focus on the home use of the device and operation of the device.
- Potential subject will read and, if wishing to participate, sign the Informed Consent Form, of which they will then receive a signed copy.
- Study staff will enroll the subject by assigning them a Subject ID and then screen the subject via:
    - Completing the inclusion / exclusion criteria Screening Questionnaire to ensure compliance with inclusion/ exclusion criteria;
    - Carrying out a finger prick blood glucose to exclude diabetes mellitus (subject will have attended fasted) (see Subject Selection Criteria);
    - Carrying out a finger prick ThyroChek test for hypothyroidism (see Subject Selection Criteria);
    - If a female subject of childbearing potential, then a urinary pregnancy test should be carried out.
- Subjects who successfully pass the screening criteria can proceed in the study and study staff will record their demographic information (age, gender, race/ethnicity), what medications (prescribed and over the counter) they take, and then randomize the subject using the block randomization method (1:1 active to control).
- Subjects will undergo measurements and assessments for Visit 1 as specified below.

- Study staff will provide the subject with the study device that they have been randomized to use throughout their home use period and give the subject training in how to use it via the reading of the instructions for use for the VeSTAL device (see Section 11 Study Device Description for details on device that will be trained to study participants).
- Study staff will also issue the subject with the following: an appropriately labeled box for the study device to stay in; sufficient electrodes and alcohol wipes to support daily usage by the subject until their next site visit; a charging cable and plug for the device; and an iPod supplied by the device sponsor to enable the subjects to use the investigational device. The iPod will be pre-installed with the VeSTAL study app that allows the device to be controlled via Bluetooth, the Activity Tracker Pedometer app, the Facebook app and a PDF in the iBooks app of the Physical Guidelines for Americans from the US Department of Health. IFU and Device Use Schematic will also be pre downloaded on supplied ipods.
- Subjects will be informed about the exercise apps that are preloaded on the iPod they are being provided with. (See Lifestyle Modification Program details described below).
- Study staff will give the subjects' registration details on how to join a slimming support group called We Slim Together (See Lifestyle Modification Program details described below).
- Dietary counseling and prescription of a hypocaloric diet with counseling on how to achieve this will be provided by a dietician. (See Lifestyle Modification Program details described below) In total 1 hour and 30 minutes will be spent with the dietician, including the time for the assessments described below.
- After completion of the various testing procedures (see below) study participants will be instructed to return home and begin using their device on a daily basis for one hour a day.

Measurements and Assessments During Visit 1

- Screening procedures (Inclusion/Exclusion criteria questionnaire; Glucose finger prick for diabetes; ThyroChek for hypothyroidism; and if applicable a urinary pregnancy test)
- Document demographic details – age, gender, race/ethnicity
- Document medications (prescribed and over the counter) that the subject takes
- Weight (Must be conducted with the participants wearing underwear and a hospital gown only – this must be consistent throughout the study duration. Weight will be taken on calibrated scales at study site, and calibration records will be reviewed during monitoring visits.)

- Height (Must be taken with shoes off and can be used to calculate BMI in conjunction with weight.)
- Visual check skin behind ears (inspect to confirm not red or broken)
- Heart rate and blood pressure
- Lipid Panel (fasting)
- Glycated hemoglobin (HbA1c)
- Fasting blood glucose
- High sensitivity C-reactive protein (CRP)
- Quality of life survey (Duke University IWQOL – http://www.qualityoflifeconsulting.com/iwqol-lite.html). Details on how to complete the questionnaires will be assessed during the SIV before study commencement.
- DXA Scan (whole body scan)
- Waist and hip measurements (To be taken in the same clothing described above, underwear and gown.) This will only be performed at the USA sites.
- Dietician to assess diet and activity via: two-day 24-hour dietary recall; completion of the EPIC-Norfolk Food Frequency Questionnaire (FFQ), and completion of the Modifiable Activity Questionnaire (MAQ).
- Hearing test/ otoscope examination of the ear canal conducted.
- Blinding assessment: As per the FDA guidance (https://www.fda.gov/downloads/medicaldevices/deviceregulationandguidance/guidancedocuments/ucm373766.pdf) the subject will be asked which intervention group (i.e. active or sham) they believe they are in and why they believe this. These data will be gathered by the VeSTAL study app, and this question will be posed to the subject immediately after they complete their first stimulation session. The subject and coordinator will not enter into discussion on this matter. These data will be passed to the study statisticians who will collate them for use in the final analyses.

Case report forms for each subject must be completed by the study team after each visit.

6.2 *Summary of Site Visit 2 (3 months)*

Measurements and Assessments During Visit 2

- Urine pregnancy test (if applicable)
- Visual check skin behind ears
- Weight (Must be conducted with the participants wearing underwear and a hospital gown only – this must be consistent throughout the study duration. They should also have an empty bladder. Weight will be taken on calibrated scales at study site, and calibration records will be reviewed during monitoring visits.)
- Completion of adverse event questionnaire by subject
- Dietician to assess diet via a two-day 24-hour dietary recall.

Other Activities During Visit 2:

- Dietary counseling from research dietician to optimize compliance with the hypocaloric diet (including review of dietary habits as above). In total one hour will be spent with the dietician.
- Ensure subject has an adequate supply of electrodes and alcohol wipes to last until next visit.
- Pay subject compensation as per compensation schedule

Case report forms for each subject must be completed by the study team after each visit.

6.3 *Summary of Site Visit 3 (6 months)*

Measurements and Assessments During Visit 3

- Urine pregnancy test (if applicable)
- Visual check skin behind ears
- Weight (Must be conducted with the participants wearing underwear and a hospital gown only – this must be consistent throughout the study duration. They should also have an empty bladder. Weight will be taken on calibrated scales at study site, and calibration records will be reviewed during monitoring visits.)
- Heart rate and blood pressure
- High sensitivity CRP
- Lipid panel (fasting)
- Fasting glucose (plasma)

- HbA1c
- Quality of life survey (Duke University IWQOL)
- Completion of adverse event questionnaire by subject
- DXA Scan (whole body)
- Waist and hip measurements (To be taken in the same clothing described above, underwear and gown.) This will only be performed at the USA sites.
- Dietician to assess diet and activity via: two-day 24-hour dietary recall; and completion of the MAQ.
- Hearing test/ otoscope examination of the ear canal will be completed.

Other Activities During Visit 3:

- Blinding assessment: As per the FDA guidance (https://www.fda.gov/downloads/medicaldevices/deviceregulationandguidance/guidancedocuments/ucm373766.pdf) the subject will be asked which intervention group (i.e. active or sham) they believe they are in and why they believe this. The subject will be asked to privately write down which type of device they believe they were given (i.e. active or sham) and also why they believe this. They subject should then fold the paper over, so the coordinator cannot see it, and pass it to the coordinator. The subject and coordinator will not enter into discussion on this matter. These data will be passed to the study statisticians who will collate them for use in the final analyses.
    - Privately (i.e. without telling the subject) the coordinator will also record which group they believe the subject to be in. The double blind will remain in place and these data will also be passed to the statisicians for analysis.
    - Document changes, if any, in medications taken (prescribed and over the counter)
    - Dietary counseling from research dietician to optimize compliance with the hypocaloric diet (including review of diet and exercise habits as above). In total one hour will be spent with the dietician at USA sites and approximately 30 minutes at UU.
    - Pay subject compensation as per compensation schedule
    - The study participants will return the investigational device and unused consumables – i.e. electrodes and alcohol wipes. They can though keep the iPod.

Case report forms for each subject must be completed by the study team after each visit.

6.4 *Schedule of Events*

Visit ID	**Visit 1**	**Visit 2**	**Visit 3**
Visit Description	**Screening/ Baseline**	**Month 3**	**Month 6 End of trial**
Visit Window (Months ± 7 Days)	**0 Months**	**3 Months**	**6 Months**
Informed Consent Form, (HIPAA & Experimental Subject's Bill of Rights for USA participants)	X		
Inclusion/Exclusion Questionnaire	X		
Glucose finger prick (Diabetes screening)	X		
ThyroChek test for hypothyroidism	X		
Urine pregnancy test (if applicable)	X	X	X
Record demographic details	X		
Record medications or change in medications	X		X
Give registration details for slimming support group	X		
Randomize into study; give allocated device and supplies; train how to use device	X		
Ensure subject has adequate electrodes/ alcohol wipes	X	X	
Check skin behind ears	X	X	X
Weight (underwear and gown)	X	X	X
Height (no shoes)	X		
Waist and hip measurements (underwear and gown)	X*		X*
Fasting glucose	X		X

Glycated hemoglobin (HbA1c)	X		X
High sensitivity CRP	X		X
Lipid panel	X		X
Heart rate and blood pressure	X		X
Quality of life survey (Duke University IWQOL)	X		X
Adverse event questionnaire	X[0]	X	X
DXA Scan (whole body scan)	X		X
Hypocaloric diet counseling	X[1]	X[1]	
Two-day 24-hour dietary recall	X[2]	X[2]	X[2]
FFQ	X[2]		
MAQ	X[2]		X[2]
Hearing test/ otoscope examination	X		X
Blinding assessment: Ask subject to indicate which intervention group they think they are in	X[3]		X[3]
Coordinator to privately record which intervention group he thinks the subject is in			X
UU site: Give subject compensation for time (see compensation schedule).			X
TDE and UCSD sites: Give subject compensation for time (see compensation schedule).		X	X
VeSTAL device and unused consumables returned to site			X[4]
Ask if subject wants to be registered for a complimentary device if FDA approval occurs			X[5]

[*] This measurement will only be performed at the USA sites.

[0] At 1-month timepoint the Adverse Event Questionnaire will be done via telephone or email. At 6-month timepoint the Adverse Event Questionnaire will be completed in person.

[1] Dietary counseling from a research dietician with emphasis on weight loss and healthy eating via a hypocaloric diet.

[2] Research dietician will have subject complete this activity.

[3] This will be performed at 0-Month and 1-Month timpoints collected via the VeSTAL app and at 6-Months in person at the clinical site. At 0-Month the VeSTAL app notification will be displayed after the first session of using the VeSTAL device

[4] If the participant completes the study to Visit 3 at 6 months then they will be given the iPod they used throughout as a gift.

[5] Staff at USA sites to ask subject if they wish their email address registered with the Sponsor so they can be notified to claim a complimentary device should medical device regulatory approval be granted as a result of this study.

Frequency of Follow-Up Appointments

As shown in the summary of frequency of follow-up appointments for all sites, the subjects will visit in person at: enrollment (0 months); 3 months and 6 months.

Telephone or email follow-up are scheduled once for the 1-month post enrollment timepoints. These follow-up sessions are estimated to take about 10 minutes each and during them the subjects should complete the adverse event questionnaire.

Also, at the 1 month timepoint the subject will be asked which intervention group (i.e. active or sham) they believe they are in and why they believe this (also referred to as a Bliniding Assessment). These data will be gathered by a pop-up question in the VeSTAL study app. The subject and coordinator will not enter into discussion on this matter. These data will be collected and analysed at the end of the study.

Study participants are also free to contact (call or email) study personnel at any time with any issues or inquiries, and there is also the facility to bring subjects in on an ad hoc basis should concerns arise.

Summary of Frequency of Folllow-Up Appointments for sites:

	Follow-up for all sites:

Clinical Site in Person Visits	Enrolement (0-Month), 3-Month and 6-Month timepoints
Adverse Events collected in Person	3-Month and 6-Month timepoints
Adverse Events collected via telephone or email	1-Month timepoint
Blinding Assessment collected via VeSTAL study application	0-Month and 1-Month timepoints (At 0-Month the VeSTAL app notification will be displayed after the first session of using the VeSTAL device.)
Blinding Assessment collected in Person	6-Month timepoint

6.5 *Summary of Dual Energy X-Ray Absorptiometry (DXA) Scanning Procedure*

Dual Energy X-ray Absorptiometry (DXA) is a technique that was originally developed to determine bone mineral density and to aid in the management of osteoporosis. More recently, the technique has been expanded to include the analysis of fat mass and lean body mass in addition to bone density. The DXA machine emits alternating high and low energy X-rays that produce precise, high quality images. A fan beam is now used, and this technology allows decreased scan times so that scans are completed within seconds or minutes.

The basic principle of DXA data acquisition is based on the differences between bone and soft tissue attenuation at the high and low X-ray levels. As the X-ray beam passes through the subject, detectors register the varying levels of X-rays that are absorbed by the anatomical structures of the subject. The raw scan data, which includes values of tissue and bone, are captured and sent to a computer. An algorithm interprets each pixel and creates an image and quantitative measurement of the bone and body tissues. The amount of radiation exposure is very low.

We will acquire a whole body DXA scans in Array mode (using the available DXA scanner at the Clinical Sites) at time-points 0 and 6 months. The technique has a precision error (1SD) of 3% for whole body fat, and automatically calculates total body fat, truncal fat and visceral adipose tissue. The radiation exposure

associated with this protocol is small, and the cumulative total the MPE indicates that the dose is 5 μSv for each DXA scan. This gives total radiation dose of 10 μSv as 2 DXA scans are performed.
10 microSieverts= 0.01 milliSieverts for the particpants.

In order for the DXA scanner to function properly all metal must be removed from the subjects' bodies and if possible they should change into a hospital gown/ light clothing. There is a DXA subject instruction sheet, this will be provided by the clinical research dietician and scanner operator at the Clinical Site, that is being included with this submission. It includes instructions like avoiding strenuous exercise and large meals and maintaining equivalent states of hydration in the hours immediately before a scan. Also, all female subjects, of child bearing potential, must undergo a urinary pregnancy test prior to each DXA scan to ensure that they are not pregnant. Due to the suggestion in the literature of a small risk of increased error, subjects will be requested to refrain from exercising (e.g. running, swimming or weight-lifting) in the 24 hours before any of the DXA scans.

6.6 *Hypocaloric Diet*

All subjects taking part in the study must agree to try to lose weight by participating in a hypocaloric diet for the duration of the study – i.e. 6 months. A hypocaloric diet is the standard lifestyle modification program used in the trials of all the FDA approved weight loss medications – i.e. orlistat (600kcal per day deficit going to 900kcal after 6 months) (Sjöström et al., 1998), lorcaserin (600kcal daily deficit) (Smith et al., 2010), phenteramine-topiramate (500kcal deficit) (Allison et al., 2011), naltrexone-bupropion (500kcal) (Apovian et al., 2013), and liraglutide (500kcal) (Wadden et al., 2013).

The subjects will receive advice and counseling on how to achieve an hypocaloric diet from the research dietician who will see the subjects for one hour of counseling and assessment at the 0, 3 and 6 month visits. At the baseline visit the dietician will calculate the subject's basal metabolic rate (BMR) by means of the Harris-Benedict equation (Harris & Benedict, 1918). The dietician will then calculate that subject's total daily energy expenditure by multiplying the BMR by 1.3 (this is the standard method followed in the weight loss drug trials as described in this paper about orlistat (Sjöström et al., 1998).

After doing this calculation the dietician will then prescribe a 600kcal deficit hypocaloric diet for each subject. The dietician will review the hypocaloric diet plan with the subject and provide them with sample meal plans

together with weight loss and weight maintenance tips. The dietician will review the subject's weight and adherence to their diet plan at the 3 month follow-up visit and provide ongoing advice and counseling.

6.7 *Summary of Dietary Questionnaires*

Dietary counselling from a professional dietician will be performed with an emphasis on weight loss and healthy eating. These counselling sessions will include the following:

1. A two-day 24-hour dietary recall at 0, 3 and 6 months
2. An EPIC-Norfolk Food Frequency Questionnaire (FFQ) at 0 months.
3. A Modifiable Activity Questionnaire (MAQ) at 0 and 6 months.

The EPIC-Norfolk Food Frequency Questionnaire and the Modifiable Activity Questionnaire will be completed manually by the participant with the dietician assisting with any questions during the dietician counselling visits or remotely were indicated on this protocol as appropriate. The two-day 24-hour dietary recall data will be analyzed via a database application called Nutritics, Nutritics will collate all two-day 24-hour dietary recall data across both sites for end analysis. This software will be trained and released to the clinical site via the sponsor at the training and Site Initiation Visit.

6.8 *Assessment of Diet and Activity*

The dietician will complete the three following assessments with each subject:

1. Dietary information will be collected using a slightly adapted version of the USDA multiple-pass 24-hour recall method, originally developed by the USDA to limit the extent of underreporting that occurs with self-reported intake. The method will be adapted to use three distinct passes and will collect information about a subject's food intake during the preceding weekday and weekend day (two-day 24-hour recall) to better reflect the overall diet. This will be done at the 0, 3 and 6 month visits.

There is no form for the subject to complete but rather involves the dietician conducting a "sweep" style interview with the subject. In brief the steps will be: (1) the quick list, which is an uninterrupted listing by the subject of foods and beverages consumed; (2) the forgotten foods list, which queries the subject on categories of foods that have been documented as frequently forgotten; and (3) the detail cycle, which elicits descriptions of foods and amounts eaten aided by the Portion Photos of Popular Foods guide ("Portion Photos of Popular

Foods" by Mary Abbott Hess, MS, RD and published by the American Dietetic Association). If possible step 1 will be undertaken by the participant at home before each research appointment. Steps 2 & 3 will be undertaken by the dietitian within the dietary consultation.

The two-day 24-hour dietary recall data are analyzed by the dietician to provide information on the total energy content consumed by the subject during the period, and the breakdown of this total in terms the percentage contributed individually by carbohydrate, fat, protein and alcohol. The dietician will use this nutrition analysis software to analyse the 24-hour dietary recall (https://www.nutritics.com/p/clinical). These data will be entered in an anonymized form into the software and dietician will be trained in advance of the study on how to enter the data.

2. EPIC-Norfolk Food Frequency Questionnaire (FFQ) which is a validated dietary research tool composed by the University of Cambridge and the UK's Medical Research Council. As it is a UK developed instrument some of the language used is particular to that country. On the advice of the FDA the FFQ has been adapted to additionally explain in American English what type of foods are being referred to. It gives a view of intake frequency for specific foods and beverages over the previous year, and so provides a validated means to compare the dietary habits of the active and sham groups at baseline. This will be assessed by the dietician at the 0 month timepoint. The dietician can also analyze the FFQ using the Nutritics software to provide a spreadsheet containing data on energy, nutrient and food group consumption for each subject.

3. Modifiable Activity Questionnaire (MAQ). This will be done at 0 and 6-months for all sites as it assesses activity over the previous year. This validated assessment of activity will provide data on: total physical activity averaged over the past year in hours per week; and total physical activity averaged over the past year in MET-hours per week. One Metabolic Equivalent of Task (MET) is a physiological measure set by convention as 3.5 ml O2·kg−1·min−1 (or 3.5ml of oxygen per kilogram of body mass per minute) and is roughly equivalent to the expenditure of 1kcal per kilogram of body weight per hour. MET value multiplied by weight in kilograms tells you calories burned per hour (MET*weight in kg = calories/hour).

When the research dietician manually enters the above data into the various assessment tools, correct data entry will be verified by the study coordinator.

6.9 *The Healthy Eating Program and Support*

Study staff will give the subjects registration details on how to join a dietary advice support group called We Slim Together (WST) (https://wstcommunity.co.uk). This is a commercial weight loss support group based in Northern Ireland that has agreed to provide a tailored weight loss support service to the trial subjects in both the USA and UK. Specifically, the purpose of this support group is to help and encourage the subjects to adhere to the hypocaloric diet prescribed by the dietician. WST follows a similar program style to Weight Watchers with dieters eating low calorie conventional foodstuffs (as opposed to shakes). A recent systematic review showed that at the one-year point there is evidence to support the efficacy of this type of weight loss program (Gudzune et al., 2015). As part of the ICF the subjects are asked to consent to the WST mentoring team being told their weight and the daily caloric target that they have been set by the research dietician. This will be done by email, and no other subject specific information will be communicated by study staff to the WST team. Moreover, the data will be attached to the subject's ID number rather than their name.

The subjects will have a weekly invitation, at a time convenient to them, for a 10-minute private discussion with a named WST mentor. In advance of this session the subject will be asked to weigh themselves and to make the mentor aware of this weight, so they can judge the subject's progress with the weight loss plan. (These weights are simply to guide this interaction will not be collected as part of the study data). Three out of every four weeks this private discussion will take place over Facebook messenger, and every fourth week it will consist of a face-to-face video call via Skype, Facetime or another analogous service. Subjects will be asked to try and attend at least 75% of these scheduled discussions with their mentor, and the WST mentors will provide the study team with monthly feedback on subject attendance.

The subjects will get access to the general WST community, which is accessed via a private Facebook group. The Facebook app will be pre-installed on the iPod. Subjects will be asked not to discuss their involvement in the trial or use of the trial device on this group, and the WST group moderators will also be aware of this injunction. Also, as part of the WST community the subjects will get access to: all WST recipes; all WST cooking videos; weekly live motivational talks; and access to 24/7 community support. Although subjects must engage with the weekly mentoring sessions if they want to take part in the study, taking part in the other aspects of the WST is left to their discretion. However, once a week the subjects will be asked by the VeSTAL app to estimate to the nearest hour what their usage of these other WST support group facilities has been.

The WST program involves encouraging participants to eat fresh rather than processed foods and educating them about low density foods that will keep them fuller for longer. The aim is to help make meals enjoyable while sticking to the hypocaloric diet set by the dietician. Thus, for instance, it will be suggested to participants that meals made up of the combinations of say 25% lean protein (with less than 5% fat), no more than 25% of carbohydrates like pasta, rice or potatoes, and about 50% vegetables.

An alternative company called Clinical Trial Mentors (CTM) may be used in substitute for WST if required i.e. if WST are unable to meet the capacity of mentoring support needed for the trial (e.g due to the impact of COVID-19). CTM provide the same service as WST and if required will follow an identical setup as described above, including the same procedures and privacy considerations.

6.10 *Exercise*

There is no mandatory exercise regimen imposed as part of the study. This is because exercise appears to be a less important factor in generating weight loss than diet (Melanson et al., 2013). This observation may in part be due to compensatory behavioral adaptations (e.g. eating more), but also the fact that out with the lower range of physical activity energy expenditure is constrained, meaning that increasing levels of exercise do not, in fact, lead to ever increasing levels of total energy expenditure. Rather, in what is termed the constrained model, total energy expenditure plateaus with increasing exercise probably due to a compensatory fall in basal metabolism (Pontzer et al., 2016).

Nonetheless, as there are multiple health benefits from being physically active, and so the subjects will be encouraged to avail themselves of the guidance and apps that are being provided to them on the iPod. The following physical activity resources are on each study iPod: the Activity Tracker Pedometer app which will allow subjects to track how many steps they take; and as a PDF in the iBooks app the Physical Activity Guidelines for Americans from the US Department of Health (see: https://health.gov/paguidelines/pdf/paguide.pdf).

No data gathered by the pedometer app will be utilized in the study. It is being provided as an aide for the subjects to use to try and get more exercise. Subjects will be told that chapters 4 to 6 and chapter 8 of the Physical Guidelines for Americans are the most informative for them to read. These are the chapters that

respectively detail the guidelines for: Active Adults; Active Older Adults; Safe and Active; and Taking Action: Increasing Physical Activity Levels of Americans.

6.11 *Impact of Weight on Quality of Life (IWQOL)*

A Quality of Life Survey (IWQOL) will be done at 0 and 6 and months by the study staff. This validated instrument, developed by Duke University, has been used by a variety of researchers to assess various obesity treatment options and their impact on individual patients. The questionnaire is available in a number of languages including Spanish. A detailed description is available on this website: https://olv.duke.edu/iwqol. As can be seen on this website this instrument has been used extensively in studies relating to weight loss and obesity: http://www.qualityoflifeconsulting.com/iwqol-litepublications.html.

6.12 *Hearing Test and Ear Examination*

It is proposed that study participants should undergo a hearing test and otoscope examination of the ear canal at 0 and 6-months “prior to and at the end of the clinical study”. The FDA has allowed a two week window after the subjects are seen and commence device usage for these tests to be carried out in. There is no suggestion from any of the literature looking at the safety of GVS that hearing, or the ear canal are adversely affected (Krystal et al., 2010; Marshall et al., 2010). And that a study specifically looking at the impact of GVS on cochlear function showed no effect (Cevette et al., 2012). Nonetheless, although there is no suggestion that hearing is adversely affected the FDA are requesting that such tests be carried out before agreeing to approve the device.

An audiogram (hearing) test will be conducted on study participants at baseline and at 6 months for all sites. Prior to the audiogram, the outer canals of the study participant’s ears will be examined with an otoscope to make sure that the tympanic membrane is intact, and that there is no infection. An audiologist will then place a pair of insert headphones (Etymotic, ER-3A) into the subject’s ears and ask them to raise a hand or press a button when they hear a soft pure-tone. Bone conduction and air conduction thresholds (dB HL) will be measured at 250, 500, 1000, 2000, 3000, 4000, 6000, and 8000 Hz bilaterally. This is a standard hearing test to determine if hearing loss is present, and if so how much and of what type. A plot of frequency versus amplitude sensitivity threshold for each ear will be generated. The speech recognition threshold and word recognition score will also be obtained. All testing will be completed in a sound-treated booth in order to avoid extraneous noise.

The clinical site will contact the hearing test provider used specific for the clinical site during the visits. For the UU site the hearing tests will be conducted at Specsavers Opticians and Audiologists, 7-8 The Diamond Coleraine, BT52 1DE. The receptionist there will be contacted by the UU Facility to arrange a suitable appointment for a full hearing assessment to be conducted at a time which is suitable for the study participant. The time and date of the appointment should be recorded on the appropriate case report from for that visit. UU, UCSD and TDE site subjects will receive an audiogram (full hearing) test at 0 and 6 months (Or assessment using the AMTAS Flex system). Prior to the audiogram, the ear canals of the study participant will be examined with an otoscope to make sure that the tympanic membrane is intact, and that there is no infection. This hearing test will be based on the standard and most common type of hearing test, pure tone audiometry. The result of the test is an audiogram diagram which plots a person's hearing sensitivity at the tested frequencies. These tests will be performed at the UCSD and the TDE clinical sites. The audiologist will return the hearing examination and test results to the clinical site on completion via email, a hard copy of the tests will be retained at the hearing clinic and collected by the sponsor monthly and the original copy along with the email filed in the patient data pack. The capture of this appointment should be reviewed by the PI or Sub PI when reviewing the patient data packs.

As an alternative to the audiology assessment outlined above it has also been agreed with the FDA that subjects' hearing may be assessed using the Grason Stadler AMTAS Flex device to assess pure tone thresholds. This is a patient self-guided device that can be overseen by the study coordinators in the clinic to assess air conduction screening and threshold audiometry. This option will only be utilized at the UC San Diego and TDE sites. It will be carried out with the subjects wearing the headphones provided with the AMTAS Flex device and in a quiet room. If any concern of an abnormal finding is raised during these investigations then the subjects will undergo audiology assessment as described above.

6.13 *Serum Assays*

Venous blood assays, with the subjects in a fasted condition, will be carried out at 0 and 6 months for glucose, HbA1c, lipid profile and high sensitivity CRP (hs-CRP). These are being analyzed as secondary (i.e. hypothesis generating) endpoints. The purpose in choosing these specific assays is to screen for change in cardiovascular risk factors. Specifically, glycemic control, lipid profile and inflammation are assessed using these tests. There is an increasing appreciation of the value of hs-CRP in assessing inflammation as a marker of cardiovascular

risk (Fonseca and Izar, 2016). The drug trials for liraglutide (Wadden et al., 2013), lorcaserin (Smith et al., 2010) and naltrexone/bupropion (Apovian et al., 2013) all demonstrated a statistically significant improvement in hs-CRP with treatment.

All subjects are provided with an iPod touch. This contains the following pre-installed applications: the VeSTAL study app that allows the device to be controlled via Bluetooth; the Activity Tracker Pedometer app; the Facebook app; and as PDFs in the iBooks app the Instructions for Use (IFU) for the study device, a Device Use Schematic and the Physical Guidelines for Americans from the US Department of Health (see: https://health.gov/paguidelines/pdf/paguide.pdf).

6.14 *Active Versus Sham Device*

Description of Device Function

The design of the sham device was inspired by the sham devices used for studies on transcranial direct current stimulation (tDCS). This is appropriate as GVS (VeNS) can be viewed as specialized variant of tDCS that targets the vestibular system rather than the cerebral cortex. Careful consideration has been given to generate a sham device with an appropriate degree of authenticity, so as to maintain subject blinding, while simultaneously not, inadvertently, causing significant vestibular system stimulation in the control subjects.

It has been observed that, due to subjects accommodating to the current, the tingling or itching sensations perceived on the skin during tDCS typically last about 30 seconds or so before abating (Nitsche et al., 2003; Paulus 2003). Based on this finding Gandiga et al. (2006) carried out a study in which both the tDCS and sham were increased in a ramp like fashion over about 10 seconds, however while the tDCS continued for 20 minutes, the sham was then turned off at the 30 second timepoint. Thus, the sham device still delivered the initial skin sensation, while at the same time not providing prolonged stimulation. The authors found that neither stroke patients nor healthy controls could distinguish between genuine tDCS and sham stimulation, even after experiencing both conditions.

The VeSTAL sham device follows this pattern by applying some stimulation to a user for a limited period of time (30 seconds), before tapering down to zero over a further 20 seconds, thus creating the impression of an active device. Moreover, subjects will have been informed in the ICF that it is normal for the sensation to be

stronger initially on first turning on the device and then for it to, quite quickly, become less noticeable. It elaborates that this is because they “accommodate” to the stimulation. The sham stimulus is applied as a positive and negative bias square wave signal to the user (just like the active device), but with a frequency of 0.8Hz.

The decision for the sham to deliver at a frequency of 0.8Hz during each stimulation session was made to reduce the likelihood of a vestibulosympathetic response, specifically as measured by muscle sympathetic nerve activity (MSNA). This is because MSNA has been directly linked to weight loss (Straznicky et al., 2012), and it would seem likely that any putative vestibular modulation of body mass composition would, at least in part, be mediated via the sympathetic nervous system. However, the 0.8Hz delivery in the sham device is less likely to affect MSNA than the 0.5Hz delivery in the active device (Grewal et al., 2009; Macefield and James, 2016).

GVS poses the additional challenge, over tDCS, when designing a sham that subjects may not only be aware of skin tingling but also of vestibular stimulation. The sham device addresses this by not only temporarily providing the sensation of skin tingling but also vestibular stimulation, without as significantly modulating MSNA (as it is at 0.8Hz rather than the 0.5Hz frequency of the active device). This should increase the likelihood that sham users believe they have been allocated an active device. Moreover, two other factors are also pertinent when considering the verisimilitude of the sham device. First, in a manner analogous to the cutaneous sensations, the perceptual salience of the vestibular sensation may decrease with time (Fitzpatrick and Day, 2004). And second, all the study subjects will be naïve to GVS.

Sham Stimulation Activity

The sham stimulation is a positive and negative bias square wave with a frequency of 0.8Hz, and (just like the active device) a maximum output current of 1mA. The stimulation is only active when the subject starts a session and lasts for a total of 50 seconds.

Sham Stimulation Activity

The sham stimulation is a positive and negative bias square wave with a frequency of 0.8Hz with a max output of 1mA. The stimulation is only active when the subject starts a session and lasts for a total of 50 seconds.

Stimulation Control / Power Output

The user can increase or decrease of the stimulation level via the mobile app or the buttons on the device. Unbeknownst to the subject the power level output is set in bands as follows:

Level 0	No stimulation applied 0mA
Level 1 – 3	Stimulation applied 0.5mA
Level 4 – 7	Stimulation applied 0.7mA
Level 8 – 10	Stimulation applied 1mA

The user has though a finer level of control when decreasing the stimulation:

Level 0	0mA
Level 1	0.1mA
Level 2	0.2mA
Level 3	0.3mA
Level 4	0.4mA
Level 5	0.5mA
Level 6	0.6mA
Level 7	0.7mA
Level 8	0.8mA
Level 9	0.9mA
Level 10	1.0mA

This level of control allows the user to decrease the stimulation if they find it too intense but maintain a level of stimulation that will give them the impression that the device is an active one.

Stimulation Time

The stimulation time is characterized by three time periods:

- Placebo Stimulation Period
- User Control Period
- Stimulation Decrease Period

The Placebo Stimulation Period is the total period of time that electrical stimulation is delivered by the device – i.e. 50 seconds. This period starts as soon as the subject increases the power level setting above zero.

The User Control Period is the period of time when the user has control over the stimulation that is being applied to them. This has a period of 30 seconds.

The Stimulation Decrease Period is the period of time when the device decreases the stimulation output to zero. This is a period of 20 seconds. During this period the rate at which the stimulation is decreased is set at one level every 4 seconds. This means that every 4 seconds the stimulation is decreased by 1 until it reaches Level 1. The device will remain at Level 1 until the 'Placebo Stimulation Period' expires.

After the Placebo Stimulation Period expires the device will no longer output any significant stimulation for the duration of the setting. The user will still see activity on the mobile app and will still be able to modify the apparent stimulation level via this app, or via the buttons on the device. When doing so the device will appear to the user to be operating as normal, including the single beep that acknowledges a change in the stimulation level (all beeps and light functions are identical between the sham and active device). However, no stimulation is delivered for the remainder of the session.

At the 12, 24, 36 and 48 minute timepoints the sham device will confirm correct electrode attachment. It will do so by delivering an imperceptible 0.03mA pulse for 10 seconds. If an issue with electrode attachment is detected with this then the app will alert the user to reposition the electrode. Only once the device has been power cycled (turned off and on again), or after pausing the delivery for more than 5 minutes, will the Placebo Stimulation Period begin again.

7. Rationale of Clinical Study

There is a growing realization that obesity can, in many ways, be viewed as a neurological disease triggered by lifestyle factors. There is clear evidence that the arcuate nucleus in the hypothalamus regulates a "set-point" for how much fat the body should have. It does so by altering appetite and metabolic rate so that deviations too far in either direction are strongly resisted. This set-point is determined by genetic, epigenetic and lifestyle factors. Thus, excessive exposure to dietary monosaccharides, such as glucose, and saturated fats, especially in childhood and adolescence, can damage the neurons of the arcuate nucleus and push the set-point up. This then can condemn sufferers to a lifetime of obesity.

Establishing a method of tuning down the set-point for body fat thus has to be a goal if we are to successfully combat the current obesity pandemic. A significant amount of animal work suggests that stimulating the vestibular system in the inner ear, by means of chronic centrifugation, actually does just that and causes a reduction in body fat. This is likely because the chronic vestibular activation is taken by the brain to represent a state of increased physical activity, and in order to optimize homeostasis it would be appropriate for the body to have a leaner physique, by reducing unnecessary energy expenditure from carrying excess fat.

It is possible to stimulate the vestibular nerve in humans by applying a small electrical current to the skin behind the ears. This is an established technology that is believed to be safe, but only previously used for research purposes. It was found in a pilot study (UCSD IRB #110538) that recurrent stimulation of this kind for two or three hours a week over four months led to a statistically significant reduction in truncal fat in the active group as opposed to the control group who underwent sham stimulation.

Given the current, and increasing levels of global obesity, it is important to determine whether non-invasive electrical vestibular nerve stimulation (VeNS), otherwise known as galvanic vestibular stimulation, is a viable treatment option, since if it were this would be of significant scientific importance. The only way to answer this question is to use human subjects.

In writing this Research Plan we have consulted the following document from the FDA, which although aimed at weight loss medications, does contain pertinent advice.
http://www.fda.gov/downloads/Drugs/.../Guidances/ucm071612.pdf

The current study will contribute to the field of knowledge about the neurological mechanisms that control body mass composition. We will also determine whether the relatively inexpensive, and non-invasive technique of VeNS is an effective treatment for obesity and fat loss. This is of particular importance given the current obesity pandemic and the serious public health challenge that obesity poses.

A potential benefit for participants is that they may experience some degree of weight loss as a result of participation. This is true for both the experimental and control groups, as both will be receiving regular weight loss counseling throughout the study from a professional research dietician, who will prescribe them a hypocaloric diet. Moreover, they will also be enrolled in a commercial healthy eating and dieting program – We Slim Together. This program will assist the subjects in making healthy dietary decisions in order to help them follow the hypocaloric diet outlined to them by the dietician, which will help with their weight loss. They will also be supplied with an iPod, which will be pre-installed with the Physical Activity Guidelines for Americans from the US Department of Health. The aim of giving the subject access to this is to encourage healthy exercise habits throughout the study.

8. Conduct of Investigation to Reduce Bias

All site staff responsible for recording and reporting results from the investigational device will be blinded to the results from previous weigh in or use of the device until the end of the study. The staff will be blinded to all previous results collected through the study of the patient records in relation to weight loss and patient records.

9. Device Description / Intended Use Statement

The proposed device is a transdermal neurostimulation product. It consists of a battery-powered headset designed to deliver low-level energy in the form of a neurostimulation waveform that modulates the activity of the vestibular cranial nerve. The delivery of this waveform is through two self-adhesive electrode pads. These pads are placed on the skin overlaying each mastoid process. When turned on, the product delivers a small electrical impulse which can be adjusted up or down by the patient.

Once adjusted to a level where the patient is aware of a tolerable, comfortable gentle swaying sensation, the stimulation delivered by the VeSTAL device should ideally remain at this level for the duration of the session, although further adjustments may be made using via a Bluetooth app or via the fixed up and down buttons on the headset or via a Bluetooth app., located just above the power button. The device can also be paused using the app by pressing the pause icon, or alternatively paused by pressing the power button on the headset twice in quick succession. When finished the patient removes the headset and disposes of the electrode pads after each use. When the product is not being used it can be charged through a micro-USB cable. For safety reasons, there are software controls in place to prevent the device delivering electrical stimulation when it is plugged into the micro-USB cable to charge the battery. When powered on, the product can be connected to an iOS or Android app via a Bluetooth Low Energy connection, which will allow fine adjustments of the neurostimulation amplitude.

The recommended use is 1 hour per day, the VeSTAL device will automatically stop stimulation if the patient reaches one hour of usage per day. The patient cannot use the device again within a 16-hour period, this is to allow some flexibility of stimulation routines if required.

10. Manufacturer

Neurovalens Ltd.

The Warehouse
7 James Street South, BT2 8DN
Belfast, U.K.

11. Device Description

11.1 *Headset Device*

Study staff will train the subject on how to use their study device. Specifically this will include: how to first prepare the skin over the mastoid processes with an alcohol wipe; a demonstration of where to place the 32mm diameter hydrogel electrodes (i.e. behind the ears on the skin overlying the mastoid processes – see Figure 1); how to access the VeSTAL study app and pair the iPod (using Bluetooth Low Energy) to the device; how to operate the device using the app; and how to use the up/down buttons on the device if unable to use the study app. Study staff will provide subjects with a printed copy of the IFU and also highlight its location as a PDF on the iPod. The Device Use Schematic will also be highlighted to the subjects – it will only be as a PDF on the iPod.

Although the 32mm diameter hydrogel electrodes are the first line electrodes to be used with the device, other stud electrodes approved for human use, can be used with the device so long as they meet the safety criterion specified by the FDA that their “maximum average power density should be less than 0.25 watts per square centimeter of electrode conductive surface area”.

Also study staff will demonstrate to the subjects: how to charge the device; to dispose of used electrodes after a stimulation session; how to store unused electrodes in a sealed manner so they do not dry out; how to turn the device on and off using the power button; and how to use the app or power button to pause a stimulation session.

Subjects will be asked to try and use their allocated device for an hour every day, and at least five times a week. Theoretically it is thought that the device may be more effective if used while sitting upright (as opposed to lying flat) (Macefield & James, 2016), due to the orientation of the otolith organs. As such subjects will be encouraged to use the device while sitting upright, and also in the evening as sometimes people can feel a bit soporific after vestibular stimulation. Subjects will be instructed not to walk around, operate machinery or drive

while using their device. Most people can tolerate up to 1.0mA without difficulty and subjects will be encouraged to turn the device up as much as they are comfortable with.

Figure 1 Trial Device Placement

Illustration of the trial device, showing the headset-style design and mastoid electrode placement.

11.2 *Software and Application*

The device incorporates embedded software and mobile application software, that is classified as a moderate level of concern according to FDA document “Guidance for the Content of Premarket Submissions for Software contained in Medical Devices” issued May 11, 2005.
Additionally, risk management information applicable to this product supports a software classification of Class A according to standard IEC 62304.

In summary, a failure of device software, resulting in hazards to which the software system can contribute, would not result in injury or damage to the health of the user or other persons. IEC 62304 requirement: to fall within Class A, this status must be valid, with or without the need for implementation of risk control measures (i.e. the SW failure may result in hazards that would not cause injury or damage; or the SW failure may result in hazards that, following implementation of risk controls, would not result in injury or damage).
The validation of software requirements will be addressed by system level testing. Test acceptance criteria and a summary of test results, along with other software and design details as required for submissions incorporating software of minor level of concern, as outlined in the FDA guidance referenced above, will be included in the future submissions for the VeSTAL device.

Within the system there are a number of stages that define how data gets from the VeSTAL device to the cloud service and subsequently the database:

1. Data logs are recorded by the study device;
2. The data logs are read from the device by the mobile application using Bluetooth Low Energy;
3. The data logs are stored by the mobile application;
4. When a full session is recorded the data logs for that session are sent to the cloud service using TLS (Transport Layer Security);
5. The cloud service parses the session data and stores it and the raw logs in the database.

These steps apply to both the active and sham devices. Along with this every record is date and time stamped at various points within the process. At point 1 a date/time stamp is generated on each log entry by the device, this is stored by the mobile application at point 3. At point 4 a start and end date/time stamp is generated for the entire session by the mobile application. At point 5 the session data is date/time stamped again to show when it was initially added to the cloud database. If any edits are performed after the data has reached the cloud service, the audit log will maintain a list of the following for each edit:

- A date/time stamp of when the change happened;
- The user that made the change;
- A snapshot of the data before the change;
- A snapshot of the data after the change.

The data recorded in this manner for each session are: duration of usage; average intensity; and average resistance encountered. During the study, study staff will only have access to duration of usage data. The app will upload these data when connected to Wi-Fi. The trial devices have storage capacity to record a few week's

worth of usage too, which they will store pending retrieval when the subjects next connect the device to the app.

In order to help the study team monitor compliance the duration of usage data will periodically be automatically assessed. This will occur once subjects have been in the trial for at least 2 weeks, and happen on a weekly basis each Monday. If a subject's usage over the previous two weeks is below 9 hours then an email alert, containing the subject's ID code and total use figure, will be sent to the study coordinators and PI. (The figure of 9 hours every 2 weeks was chosen to allow a clear margin below what the subjects are asked to achieve as a minimum). The study team can then monitor the situation and judge how to proceed to optimize compliance. This may include contacting the subject to encourage device usage and asking if anything can be done to help with the process.

Subjects have the option, if they so wish, to input their weight into the study app, which will display to the subjects a record of the entered values over time. This is purely for the subject's own information, and to assist their We Slim Together (WST) mentor in best advising them how to follow their prescribed hypocaloric diet to lose weight. The WST mentor will be able to view the entered weight values (attached to the subject's ID number and devoid of any subject identifiable data) via a password-accessible database. No other information from the app, other than the weight that the subject chooses to enter, will be made available to the WST team. Conversely, weight values entered into the app by the subjects will not be accessible at any point by the study staff and have no role in any of the endpoint analyses.

11.3 *Associated Devices, Consumables*

The *VeSTAL* device as part of a system, using consumable electrode pads and wipes, Neurovalens should be contacted to provide additional consumables if required throughout the trial.

All subjects are provided with an iPod touch. This contains the following pre-installed applications: the VeSTAL study app that allows the device to be controlled via Bluetooth; the Activity Tracker Pedometer app; the Facebook app; and as a PDF in the iBooks app the Physical Guidelines for Americans from the US Department of Health (see: https://health.gov/paguidelines/pdf/paguide.pdf).

Any other material/equipment required for testing are provided/sourced by the investigation site directly.

12. Instructions for Use

IFU is included as a supporting document to this protocol.

13. Packaging and Labelling

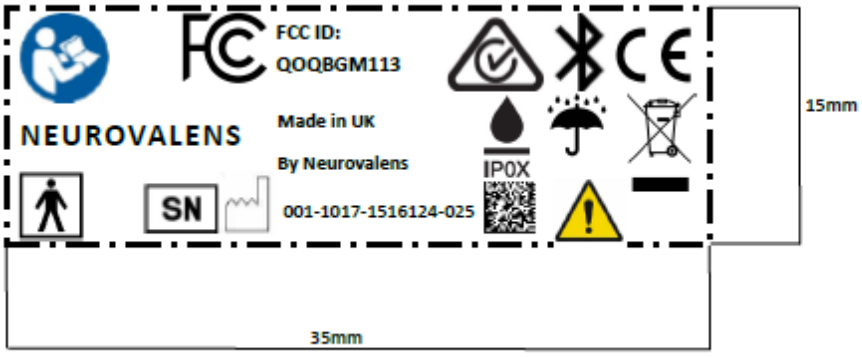
The VeSTAL Device is delivered with the following components:

- Alcohol wipes
- Electrode pads
- USB charging cable

Each of these components is labelled with the following information:

- Title
- Date of Manufacture
- Stated volume
- Lot number
- Storage conditions

The Headset Labelling is as follows:



The Headset Labelling is as follows for the USA:

"Caution: **Investigational device**. Limited by Federal (or US) law to investigational use"

Batch Number:

Serial Number:

Outer packaging USA:

CAUTION--Investigational device. Limited by Federal (or United States) law to investigational use. Vestibular Stimulation to Trigger Weight Loss (VESTAL) Clinical Trial. Clinical trial number: NCT03640286.

Manufacturer: Neurovalens Ltd, Floor 4 The Warehouse, Belfast, BT2 8DN

Outer packaging EU:

Manufacturer: Neurovalens Ltd, Floor 4 The Warehouse, Belfast, BT2 8DN

Warnings and Precautions:

- Discontinue use and contact study personnel if unexpected reactions occur.
- Possible adverse effects include: skin irritation; discomfort from the stimulation; sensation of feeling off-balance; occasional nausea; headache; and rarely vomiting.
- Only apply to the mastoids (the hard-bony area behind your ear).
- Use only while seated or lying down.
- Do not use while mobile (moving), as it can affect your sense of balance.
- Do not use while driving, or operating machinery of any kind.
- Do not use if you have been diagnosed with epilepsy.
- Do not use in the presence of a pacemaker, implanted defibrillator or neurological stimulation device (e.g. deep brain stimulation or vagal nerve stimulation device).
- Do not use in wet environments.
- Do not share your headset with anyone else.
- Do not try to remove the batteries. They are non-user replaceable.
- Keep away from children. Small items such as electrode pads are choking hazards.
- If you feel discomfort during use, pause and reduce intensity before resuming.

Contents: 1 VeSTAL device; 1 micro USB charger; 1 USB plug; 180 x hydrogel electrodes; and 200 alcohol wipes.

Made in UK. Neurovalens, Belfast, UK.
Contains 1 x VeSTAL headset, 1 x charging USB cable, 1 x box of alcohol wipes (100), 2 x bags of electrode pads (100)

"Must not be used while driving or operating machinery of any kind. Should not be used in the presence of an implanted cardiac pacemaker, defibrillator or neurological stimulation device. Possible hazards or adverse effects include: skin irritation; discomfort from the stimulation; sensation of feeling off-balance; occasional nausea; and rarely vomiting.
Warnings and precautions: it is recommended that you use this device only while seated or lying down."

14. Storage and Accountability

14.1 *Accountability*

Investigational devices will be supplied by Neurovalens. Study Staff will be required to record all material received, used and destroyed to ensure full accountability. Returned devices will be evaluated for operational compliance at the end of the study.

14.2 *Storage*

The information supplied with the VeSTAL devices state to keep them dry and out of direct sunlight. There is no other special storage instruction which accompany the VeSTAL device.

15. Device Training

All investigation site staff will be trained to the relevant level of investigation conduct in device usage. Training will include:

- Instructions on the use of the device
- Health and Safety aspects
- Clinical Investigation Plan and procedures there-in

- Completion of investigation documentation
- Handling/reporting complaints and adverse events

Training records will be maintained for all site staff involved in the study for audit purposes.

16. Risk Benefit Analysis

The risks to participants in this study are relatively small (skin irritation, mild discomfort, possible nausea, headache and, compared to background, a very small dose of radiation), as well as transient in nature; the discomfort and nausea will stop soon after stopping VeNS. And headaches have been observed to be mild and self-limiting or relieved by simple analgesia. Conversely, in terms of the importance of the knowledge that a positive study would yield, the potential benefits are significant. The global obesity pandemic poses significant public health challenges and developing novel strategies to combat it are of paramount importance.

The current study explores the possibility that the non-invasive and relatively inexpensive technique of VeNS may be useful as a weight loss adjunct. Given the importance of the knowledge that may be gained, we believe the small risks to the subjects are reasonable.

In the line with ISO 14971 a risk analysis has been conducted. The majority of risk has been mitigated through product design and manufacturing e.g. consideration was given to the risks associated with biocompatibility and safety etc. and therefore materials such as phthalates have been eliminated from the devices manufacture. The residual risk to patients who are administered this intervention is low. A list of potential risks associated with the device, procedures undertaken to minimize them, and methods used for their management are shown below.

16.1 *General Risks*

1. Information of a personal nature will be collected in order to determine eligibility for the study, and therefore there is a risk of loss confidentiality.
2. Subjects may experience some degree of distress or anxiety due to confusion, the personal nature of the questions, the usage of the trial devices, or their disqualification based on the exclusion criteria.

Risks Relating to Screening Tests

1. A diagnosis of diabetes mellitus may be suggested.
2. A diagnosis of hypothyroidism may be suggested.
3. A diagnosis of pregnancy may be suggested.

Risks Relating to Phlebotomy

1. Pain
2. Fainting
3. Bruising
4. Rarely, a small hematoma or infection where the needle punctures the skin
5. The tourniquet may cause discomfort or bruising of the upper arm

Risks related to VeNS

A risk assessment for the device has been carried out in accordance with ISO: 14971: 2012 and is held on site at Neurovalens, Neurovalens have submitted a risk request for the VeSTAL device for the study site located in USA, which indicates that it is a non-significant risk device determination. Due to the low current (1mA) and voltage (4.25 V), the VeNS stimulator being used in the study is safe. However, the following specific risks do exist:

1. Skin irritation at the electrode sites.
2. That the stimulation sites may be uncomfortable at the time of stimulation – an electrical tingling sensation may occur and also VeNS may induce the sensation of being pushed towards the side of the cathode.
3. A sensation of disequilibrium, analogous to being on a boat, may occur during the test that some subjects find uncomfortable.
4. VeNS may induce nausea.

5. VeNS may occasionally induce vomiting.
6. Headache

Risks Related to Being Weighed

Subjects must change into a hospital gown prior to being weighed, which some subjects may find embarrassing or uncomfortable.

Risks related to DXA Scanning

1. For the site based in the USA it has been calculated that subjects will be exposed to a combined total radiation dose of 0.09000mSv from the two whole body scans they will receive over the course of participating in the study. The scanner used at the clinical sites in the USA is a Hologic Horizon W DXA scanner, and the scans administered will be whole body scans in Array mode. Ionizing radiation is known to carry a dose related increased risk of cancer.
2. For the site based in Northern Ireland it has been calculated that subjects will be exposed to a combined total radiation dose of GE Lunar iDXA system (approx. 0.0025mSv from 2 whole body scans). The MPE indicates that the dose is 5 µSv for each DXA scan. This gives total radiation dose of 10 µSv as 2 DXA scans are performed.
   10 microSieverts= 0.01 milliSieverts

The scan sequences we are proposing to use were selected to provide the greatest possible information about body mass composition while exposing the subjects to the lowest possible dose of ionizing radiation.

3. In order for the DXA scanner to function properly all metal must be removed from the subjects' bodies and they must change into a hospital gown. Some subjects may find doing so to be embarrassing or uncomfortable.

Risks relating to Weight Loss Programs

1. Subjects will have to discuss their dietary habits and weight loss aspirations with a professional dietician and some subjects may find doing so to be embarrassing or awkward. Also, as with any personal data there is a risk of loss of confidentiality.
2. Data pertaining to the subjects' diet and physical activity will be collected via interview with the dietician at the baseline, 3 and 6 month visit. As with any personal data there is a risk of loss of confidentiality.
3. Subjects will also liaise with mentors from the dieting group We Slim Together, and some may be uncomfortable about doing so.

Risks Relating to Audiograms

1. Some subjects may find the process of otoscope examination of the ear canal prior to the audiogram to be uncomfortable.
2. It is theoretically possible that the otoscope examination could cause some local trauma to the ear canal.
3. It is possible that the audiogram test may reveal some hearing loss that the subject was not previously aware of and that this could cause them psychological distress.
4. Minor discomfort or pressure may be experienced from headphones.

Risk of Loss of Confidentiality

This is a general risk that applies to all of the data gathered as part of the study. The processes for managing the risk of loss of confidentiality are described in Section 39.

16.2 *Risk Management Procedures*

The risks to subjects participating in this clinical investigation are further reduced by the following factors:

- Subjects will only be enrolled if they meet inclusion criteria
- Investigators will be selected based upon their knowledge and expertise in the field and will be trained to use the device.
- Clear instructional material will be provided with each device.
- Subjects will be closely followed by trained investigators and their clinical research staff to monitor safety.
- Procedures performed in the study are established and will be administered by trained and qualified clinical research staff.

Risks relating to Screen Visit

1. If a diagnosis of diabetes mellitus is suggested during screening, then the subject will not be able to continue in the study. Instead the subject will be advised that this is a possibility and that they should see their primary care physician for further assessment regarding this possible diagnosis. Some subjects may not be aware that they have diabetes, but if it is diagnosed it allows treatment to be undertaken which can significantly reduce the risk of the various ill effects that diabetes can cause.

2. If a diagnosis of hypothyroidism is suggested during screening, then the subject will not be able to continue in the study. Instead the subject will be advised that this is a possibility and that they should see their primary care physician for further assessment regarding this possible diagnosis. Some subjects may not be aware that they have hypothyroidism, but if it is diagnosed it allows treatment to mitigate its adverse effects.
3. If a diagnosis of pregnancy is suggested during any of the pregnancy screening tests, then the subject will not be able to continue in the study. Instead the subject will be advised that this is a possibility and that they should see their primary care physician for further assessment. Some subjects may not be aware that they are pregnant, but if so then it is important that this is something they are made aware of.

Risks Relating to Phlebotomy

1. Phlebotomy will only be carried out by clinical site staff who are trained in the procedure.
2. Subjects will be seated at the time of the procedure to mitigate risk in case they faint.
3. The phlebotomist will apply local pressure to the site after the blood draw.
4. It will be confirmed that bleeding has stopped and then an appropriate bandage will be applied.

Risks Related to Vestibular Nerve Stimulation (VeNS)

1. The devices being used have been judged to be non-significant risk devices in the risk assessment, carried out according to FDA criteria.
2. Any potential subjects a history of irritation to the skin behind the ear will be excluded (See Criterion 3 in Exclusion Criteria).
3. Hypoallergenic hydrogel electrodes of 32mm diameter will be used in order to minimize the likelihood of skin irritation. If the subjects try these electrodes and do not like them then any alternative electrode they are offered will comply with the stipulation from the FDA that the “maximum average power density should be less than 0.25 watts per square centimeter of electrode conductive surface area”.
4. If skin irritation occurs after using the device the subjects will be advised to apply aloe vera or other moisturizing gel, and they will also be given a contact number at for the appropriate clinical site, UU, TDE or UCSD and they can call in order to arrange a review.
5. Prior to the beginning of the study the possible discomforts of VeNS will be described to the participant in lay language, and the investigator will make sure that the subject understands what will happen when using the device.

6. The usage of the device is electronically limited to just 60 minutes per day.
7. Usage is voluntary, and subjects will be informed at the beginning of the study that they may withdraw at any time, for any reason, with no negative repercussions.
8. Subjects will be warned about the sensation of disequilibrium and will be advised that the device should be used only while seated.
9. The maximum current the device can deliver is 1mA but there is no requirement for the subjects to use it as this level. Instead they will be asked to increase the current (using buttons on the device) to a level that is comfortable. The minimum setting is 0.1mA and the current can be increased in 0.1mA increments.
10. If subjects find the sensation invoked at the level they have the current set, to be uncomfortable (e.g. in terms of nausea or disequilibrium) then they can lower the stimulation level to a more comfortable setting.
11. The subjects can always take off the devices at any point they wish.

Risks Relating to Being Weighed

A private area will be provided for the subjects to change into a hospital gown.

Risks Related to DXA Scan

1. All female participants will have to provide a negative urinary pregnancy test prior to DXA scanning due to the very small amount of ionizing radiation involved in DXA scanning.
2. If participants become claustrophobic during the scanning process, the scan will be stopped, and the participant will be able to leave the room. Given the scans can be conducted in around 5 minutes this risk should be minimal.
3. Thorough prescreening of all patients prior to the DXA scan to make sure they have no metal on their bodies or clothing. Participants will also be asked to wear a hospital gown, which will be available to them at the clinical site.
4. They will be provided with a private area to change back and forth from their clothes into the gown.
5. The dose of ionizing radiation from these scans is very small (it has been calculated that it will add up to a total of 0.09000mSv for the two whole body scans in the USA and the MPE indicates that the dose is 5 µSv for each DXA scan. This gives total radiation dose of 10 µSv as 2 DXA scans are performed, 10 microSieverts= 0.01 milliSieverts in the UK. Exposure to this small dose of radiation will be explicitly mentioned in the consent document.

Risks pertaining to Weight Loss Programs

1. The Dietician at the clinical investigation sites who will administer the weight loss advice will be a qualified professional, employed by the clinical site with experience of such studies in the past.
2. We Slim Together is a professional weight loss group. The only data that will be shared with them will be done with each subject's consent and will consist of the hypocaloric dietary target set by the study dietician, the subject's weight and an ID number for the subject.
3. Subjects will be free, if they so wish, to use a pseudonym while interacting with WST.

Risks Relating to Audiograms

1. The examinations will only be carried out by trained clinical professionals.
2. Participants will be free at any time to indicate that they wish to stop the procedure.
3. The risk of revealing a previously unknown hearing impairment will be described in the Informed Consent Form.
4. If headphones are not tolerated, then a different type of ear phone will be employed, which consist of a soft foam which is placed at the entrance to the ear canal.

17. Scope and Objectives

17.1 *Primary Objective*

To establish the clinical performance of the VeSTAL Device stimulation effect in patients who are overweight.

17.2 *Secondary Objective*

To evaluate the safety of the VeSTAL device relative to control group, in terms of the occurrence of adverse events, changes from baseline in vital signs (blood pressure and heart rate), and safety laboratory results (chemistry and haematology), in patients who are overweight.

17.3 *Other objectives*

Evaluate the representativeness of the study population in terms of the balance in demographic and disease

state covariates.

18. Overall Design

18.1 *Endpoints for USA Regulatory Clearance*

Primary Endpoints USA:

The efficacy of the VeSTAL device within the treatment group will be quantified by calculation of two primary endpoints after 6 months of treatment:

- mean percent weight loss;
- proportion of participants who lose at least 5% body weight.

Study Acceptance Criteria USA:

- Mean: There should be at least a 2% total body weight loss (TBWL) superiority margin between the active-product and sham-treated groups.
- Categorical: The proportion of participants who lose 5% TBWL or more in the active-product group is at least 50%, independent of the sham control

Both weight loss efficacy acceptance criteria must be met to claim success. These criteria can be specified in mathematical formulae as follows:

Rejection of both the null hypotheses $\mu A - \mu C \leq 2\%$ and $\pi A \leq 50\%$ at the 2.5% (1-sided) level

Where:

μA and μC are the underlying population mean percentage weight loss on active and control respectively

πA is the underlying population proportion of subjects who lose ≥5% of baseline body weight on active.

18.2 *Endpoints for EU Regulatory Clearance*

Primary Endpoint EU:

The efficacy of the VeSTAL device within the treatment group will be quantified by calculation of the following endpoint:

1. Mean percent weight loss

Study Acceptance Criterion EU:

- Mean: There should be at least a 2% total body weight loss (TBWL) superiority margin between the active-product and sham-treated groups.

This weight loss efficacy acceptance criterion must be met to claim success. This criterion can be specified in mathematical formulae as follows:

$mA - mC \geq 2\%$ and $pt \leq 0.05$

Where:

mA and mC are the observed mean percentage weight loss on active and control respectively

pt is the p-value (two-sided) from a linear regression comparing percentage weight loss between active and control ($H0: \mu A = \mu C$; $H1: \mu A \neq \mu C$)

μA and μC are the underlying population mean percentage weight loss on active and control respectively

18.3 *Other Endpoints for USA and EU Regulatory Puposes*

<u>Secondary Endpoints which may become the basis of claims in USA and EU (At 6 months):</u>

Total body fat loss (as measured by whole body DXA scanning), dietary outcomes (as measured by the two-day 24-hour dietary recall), quality of life measures (as measured by the IWQoL questionnaire) and blood parameters are secondary endpoints which may become the basis of labelling claims. As such they will be tested hierarchically after the primary endpoints (full details of the hierarchical testing procedure will be detailed in the Statistical Analysis Plan).

Secondary Outcome Measures USA and EU (At 6 months):

- The difference in mean fat mass loss (in grams) between the active-product and sham-treated groups is statistically significant. (As measured by means of a whole body DXA scan).
- The difference in mean trunk fat loss (in grams) between the active-product and sham-treated groups is statistically significant. (As measured by means of a whole body DXA scan).
- The difference in mean visceral adipose tissue (VAT) mass loss (in grams) between the active-produce and sham-treated groups is statistically significant. (As measured by the whole body DXA scan).
- Difference in mean lean mass gain (in grams) between the active-product and sham-treated groups is statistically significant. (As measured by means of a whole body DXA scan).
- Difference in mean bone mineral content (BMC) gain (in grams) between the active-product and sham-treated groups is statistically significant. (As measured by means of a whole body DXA scan).
- Fasting glucose
- Lipid profile (Assess total non-HDL cholesterol, and HDL to total cholesterol ratio)
- High-sensitivity C-reactive protein
- Glycated hemoglobin (HbA1c)
- Two-day 24-hour dietary recall
- Modifiable activity questionnaire
- Blood pressure
- Heart rate
- Waist and hip circumferences (This will only be performed at the USA sites.)
- Body Mass Index (BMI)
- Quality of life – IWQOL-Lite questionnaire.
- Dose response analysis: it is likely that some subjects will not utilize their device for the mandated 7 hours per week. Usage data will be available from the devices permitting a dose response analysis, which will done be in an intention to treat manner.
- Adjustment of medication – both reduction and new medications.
- Mentor support group usage (total hours).

Safety Endpoints

<u>Adverse Event Monitoring Questionnaire (At 1, 3, and 6-months)</u>

Based on the survey of adverse events devised by Utz et al. (2011) occurrence of the following adverse events will be specifically enquired about by having the subjects complete the Adverse Event questionnaire: headache; pain/irritation behind the ears; vertigo/ dizziness; blurred vision; nausea; fatigue/ tiredness; seizures; tinnitus; and other symptoms. This will be done in person during the 3 and 6 months in person and at the 1-month timepoint, this questionnaire will be conducted via telephone or by email. It will also be completed on an ad hoc basis should the need arise. The questionnaire will ascertain if any of these symptoms occur during or after use of the device, and if so how frequently.

We will monitor each instance of an adverse event causing a dropout from the study. The dropouts, together with causes, will be recorded and reviewed at the end of the study. This guidance from the FDA will be followed: https://www.fda.gov/downloads/RegulatoryInformation/Guidances/UCM126572.pdf. If it becomes evident that the proportion of subjects who drop out due to safety concerns/ adverse events convincingly exceeds 30%, the study will be halted.

19. Number of Subjects and Investigational Sites

An anticipated number of 200 patients shall participate in the device investigation. Subject enrollment will take place across two sites:

1. University of California San Diego (UCSD)
2. University of Ulster (UU)
3. Texas Diabetes and Endocrinology (TDE) site

20. Subject Selection

20.1 *Target Population*

Patients who are overweight.

20.2 *Subject Selection*

Pre-enrollment Screening

Prior to being allowed to start the study proper, potential subjects with type 2 diabetes should be excluded by means of a fasting blood glucose test. This can be via a finger prick sample and glucose meter. Testing should

be done in the morning after an overnight fast of at least 8 hours. If the reading is 126 mg/dl or above, then the potential subject will be excluded from the study and asked to see their primary care physician in order to be investigated for type 2 diabetes mellitus. At the same Screen Visit subjects will complete a questionnaire about their medical history and medications to ensure that they fulfil the inclusion and exclusion criteria for the study. Any concomitant medications taken by the subject will be captured via the Concomitant Medication Form. The questionnaire will be identified using an alphanumeric code and will not have any identifiable subject data on them. The source documents will be securely stored at the Clinical Site.

The following procedures will also be performed during the Screening process:

- Obtain Informed Consent
- Obtain demographic data, including gender, date of birth/age, race, and ethnicity;
- Screening questionniare to confirm conformity with inclusion and exclusion criteria – access to medical records not required, medical history collected via questionnaire.
- Measurement of height and weight to allow BMI to be calculated
- Pregnancy test on urine (for women of childbearing potential only)
- Hypothyroidism screening test
- Fingerprick blood glucose

20.3 *Subject Recruitment*

The clinical sites at UCSD) will recruit subjects via Research Match (see: https://actri.ucsd.edu/clinical/Pages/using-ResearchMatch.aspx). This is a “is a NIH-sponsored national registry of volunteers who have indicated a willingness to learn more about research studies.” The aim of this registry is to connect “researchers with appropriate potential subjects.” The UCSD and TDE sites will also recruit via social media advertizing (e.g. Facebook), social networking advertizing (e.g. Craigslist), newspaper adverts, posters, leaflets, tabling events, use of volunteer databases and potentially radio advertizing.

For the TDE site, as they are a clinical practice with an internal research centre, they will also recruit subjects via their patient database. Existing patients may be called or sent emails with the relevant study information or through social media or network advetrtising as well.

For the clinical trial site based in Northern Ireland potential subjects will be targeted via social media campaigns such as Facebook, Twitter and Instagram as well as radio and local news advertising.

In order to improve recruitment numbers, we also plan to post recruitment flyers in the hospitals and universities associated to where the clinical trial is taking place. The advertisement to Facebook groups relating to weight loss that operate in the local areas where the clinical sites are located; if necessary Neurovalens may ask Facebook to promote this statement via a paid advertisement.
Neurovalens shall be responsible for recruitment and shall follow best practice for the management of bias in subject recruitment.

20.4 *Inclusion Criteria*

1. Signed informed consent
2. Body mass index BMI ≥ 27 kg/m2.
3. Males or Females. Note females of child-bearing potential must have a negative urine pregnancy test at screen and also just before each DXA scan. (As DXA involves a small dose of ionizing radiation). They should agree to follow a physician-approved contraceptive regimen for the duration of the study period (other than DMPA injections as this causes weight gain).
4. 22-80 years of age inclusive on starting the study. (In order to comply with FDA guidance: https://www.fda.gov/MedicalDevices/DeviceRegulationandGuidance/GuidanceDocuments/ucm089740.htm#s6)
5. Ability and willingness to complete all study visits and procedures; in particular an agreement to engage with: trying to use the device on a daily basis; the hypocaloric diet weight loss program; and this provided weight loss support and mentoring.
6. Agreement not to use of prescription, or over-the-counter, weight loss preparations for the duration of the trial.
7. Agreement not to start smoking tobacco or marijuana for the duration of the study.
8. Access to Wi-Fi (to connect iPod to internet)

20.5 *Exclusion Criteria*

1. History of vestibular dysfunction or other inner ear disease as indicated by the screening questions.
2. History of bariatric surgery, or gastric resection.

3. History of skin breakdown, eczema or other dermatological condition (e.g. psoriasis) affecting the skin behind the ears.
4. History of weight loss device implantation (e.g. VBloc Maestro or Abiliti).
5. Use of a non-invasive weight loss device (e.g. Modius)
6. Hypothyroidism requiring current treatment with levothyroxine (e.g. Levo-T, Synthroid, Thyroxine) (Other thyroid disorder patients on stable treatment for at least 3 months are acceptable).
7. Other endocrinological causes of weight gain (e.g. Cushing's disease, Cushing's syndrome or acromegaly)
8. Previous diagnosis of HIV infection or AIDS (HIV is known to cause a vestibular neuropathy which would prevent VeNS from working).
9. Diagnosis of cirrhosis, chronic pancreatitis, or liver, kidney or heart failure.
10. Treatment with prescription weight-loss drug therapy in the 6 months before starting the study.
11. Tobacco smoking (including vaping) in the six months prior to starting and for the duration of the study.
12. Use of marijuana (smoking, vaping or in edible form) more than twice a month on average.
13. Known genetic cause of obesity (e.g., Prader-Willi Syndrome).
14. Body weight change of more than 20% in either direction within the previous year.
15. Physician-prescribed diet, and/ or current, active member of an organized weight loss program.
16. Diabetes mellitus (Types 1 & 2).
17. Diagnosis of epilepsy or use of anti-epileptic medication within six months of starting the study (e.g. for the treatment of peripheral neuropathy)
18. Chronic (more than a month of daily use) treatment with opioid analgesic drugs within the last 6 months.
19. Regular use (more than twice a month) of anti-histamine medication within the last 6 months.
20. Use of oral or intravenous corticosteroid medication within 6 months of starting the study.
21. Use of the beta-blockers atenolol, metoprolol or propranolol within 3 months of starting the study.
22. Current alterations in treatment regimens of anti-depressant medication for whatever reason (including tricyclic antidepressants) (Note: stable treatment regimen for prior 6 months acceptable).
23. An active diagnosis of cancer.
24. A myocardial infarction within the preceding year.
25. A history of stroke or severe head injury (as defined by a head injury that required craniotomy or endotracheal intubation). (In case this damaged the neurological pathways involved in vestibular stimulation).
26. Presence of permanently implanted battery powered medical device or stimulator (e.g., pacemaker, implanted defibrillator, deep brain stimulator, vagal nerve stimulator etc.).

27. Psychiatric disorders (including untreated severe depression, schizophrenia, substance abuse, eating disorder etc.)
28. Current participant in another weight loss study or other clinical trial.
29. Have a family member who is currently participating or is planning to participate in this study.
30. Weight over 350 pounds at UU, TDE and CTRI site (as this is the weight limit of the DXA scanner) or a weight over 500 pounds at the EPARC site (as this is the weight limit of the DXA scanner)
31. Pregnancy

The decision to exclude participants with type 2 diabetes mellitus was made after reading the FDA's draft guidance (see: http://www.fda.gov/downloads/Drugs/.../Guidances/ucm071612.pdf). This notes that, "[c]compared with nondiabetic patients, overweight and obese patients with type 2 diabetes often respond less favorably to weight-management products and may face unique safety issues such as risk for sulfonylurea-induced hypoglycemia following weight loss (if the dose of sulfonylurea is not appropriately lowered or the drug discontinued)."

The FDA then go on to advise "examining the efficacy and safety of weight-management products in trials dedicated to patients with type 2 diabetes." On the basis of this advice we made the decision to exclude patients with type 2 diabetes. This is an important patient group of course, and our intention is to investigate type 2 diabetics in due course.

If a subject is found to possibly have type 2 diabetes during the initial screening, then they will be excluded and asked to notify their primary care physician. However, if a subject passes the initial screening but a suspicion of this diagnosis arises during one of the study tests, then the subject will be asked to notify their primary care physician, though they will still be allowed to complete the study.

20.6 *Withdrawal criteria*

These processes are to be implemented at both study sites.

Subjects who do not meet the screening criteria will be considered screen failures and will be replaced.

Subjects may withdraw from the investigation for any reason, at any time. Subjects are free to participate or not participate as they wish.

During the course of the study, subjects may be withdrawn from further involvement. Reasons for withdrawal may include:

- Subject removal of consent
- Any adverse event (related or not) that in the opinion of the investigator will endanger the well-being of the subject if they continued in the study.
- Poor compliance with the study requirements (protocol violation)
- Subjects lost to follow-up

All subjects withdrawn from the study due to unexpected device effect, directly related to the clinical investigation, will be followed-up and treated until the effect resolves.

Subjects who are withdrawn will not be replaced. All discontinuations will be documented along with the reason for withdrawal. Reasonable efforts will be made to contact any subject lost to follow-up during the course of the investigation in order to complete assessments and retrieve data. Should a subject be lost to follow-up the data available will be used in the final report, all uses of partial data will be documented. Ethics Committees and regulatory authorities will be notified of withdrawal as required by the regulations.

If possible, when a subject withdraws, or is withdrawn, a final visit should be conducted to allow a withdrawal discussion with the subject. At this visit the study device, iPod and any unused consumables should be collected. Also, at this visit it will be requested in a non-coercive manner if the subject will permit their weight to be taken, and if they are willing to complete the Adverse Event monitoring questionnaire. Moreover, if the subject had reached the point where they were due a follow-up appointment (i.e. at 3 and 6 months), then they be asked, again in a non-coercive manner, whether they are willing to have the scheduled tests and measurements for that appointment. It will though be made clear to the subject that these activities are optional on their part.

If the subject does not wish to return for a final visit, then a withdrawal discussion on the telephone will be attempted. In this discussion factual information may be provided to the subject and arrangements made for end-of-study activities like return of study investigational device, payment of any outstanding compensation

and whether the subject will complete the adverse event questionnaire over the phone. There will be no attempt to coerce or unduly influence the subject.

A list will be assembled of all subjects who withdrew, or were withdrawn, from the study after enrollment. This will be broken down by center and treatment group and contain the following information: subject ID; specific reason for discontinuation; duration of treatment and last day of study adherence before discontinuation; whether or not the blind for the subject was broken at the time of discontinuation; if the subject returned the study device; and, after conclusion of the study and unblinding, what that subject's treatment allocation was – i.e. active device or sham.

21. Investigation Procedures

21.1 *Informed Consent*

In order to facilitate the identification of prospective research participants who meet the eligibility criteria for enrollment review it is proposed that pre-screening by telephone take place. In order to allow this to take place over the telephone it is requested that a waiver of consent documentation is allowed.

As this pre-screening process will collect identifiable information that includes Protected Health Information (PHI) prior to obtaining consent to participate in the study, we wish to request a waiver of individual HIPAA authorization. Below we describe how each of the following criteria for granting the waiver will be satisfied:

a) Why the disclosure of PHI involves no more than minimal risk.

This process is considered minimal risk to the potential subjects as all data gathered via this process will be treated in the same confidential manner as data gathered at any other part of the study. Moreover, all data for subjects deemed ineligible to participate will be destroyed at the end of the phone call to pre-screen them.

b) Why granting the waiver will not adversely affect privacy rights and welfare of the individuals whose records will be used.

It is not believed that this process will adversely affect the privacy rights and welfare of potential subjects that are pre-screened as the telephone script still includes the Elements of Consent. Potential subjects are thus

requested to give verbal consent to undergo the pre-screening process. Other than the data provided over the phone by potential subjects undergoing the pre-screening process no hospital records or other PHI will be accessed as part of the telephone pre-screening process.

c) Why the project could not practically be conducted without a waiver.

The pre-screening process takes place over the phone it is not possible to obtain signed consent.

Without the waivers of documented consent and individual HIPAA authorization in place the recruitment process would be much more cumbersome, a lot less practical and potentially significantly inconvenient for the potential subjects, as there are multiple inclusion and exclusion criteria. Thus, many potential subjects would attend in person at Clinical Site despite being clearly ineligible to participate in the study.

d) Why the project could not practically be conducted without the use of PHI.

There are over 30 inclusion and exclusion criteria, many of which pertain to aspects of PHI, and several of which relate to various relatively common health conditions and medications. Thus, without the use of PHI it would be impossible to inadequately screen potential subjects properly, and many would attend in person at the Clinical Site despite being obviously ineligible to participate in the study.

e) An adequate plan to protect identifiers from improper use and disclosure.

No identifiers will be attached to the pre-screening documentation, and no identifiers will be collected during the pre-screening process. At the end of the phone call to pre-screen subjects it will be determined whether the subject appears likely to be eligible or ineligible to pass the study's screening process. Eligible subjects will be invited to attend the Clinical Site to be consented and enrolled into the study. Once this determination regarding eligibility is made the pre-screening questionnaires will be destroyed for all subjects.

A separate document detailing the potential subjects who have been contacted via the pre-screening process will be kept in a locked data cabinet in the Clinical Site. (If this information is stored on a computer, then it will be on a Clinical Site computer assigned to study staff by the Clinical Site for research purposes, and it will require a password for access, which only personnel directly involved in the study will have.) No PHI will be on this document. Rather, all that will be recorded is whether the potential subject was found eligible or ineligible

via the pre-screening process. (It is necessary to keep a record of the names of the potential subjects found ineligible to proceed to avoid accidentally contacting the same person twice during the recruitment period).

Access to these data in any format will only be available for study staff on the protocol. All data gathered via this process will be treated in the same confidential manner as data gathered at any other part of the study. Once recruitment to the study is complete (i.e. 200 subjects have successfully passed the screening process after giving informed consent) then this list of subjects who underwent the pre-screening process will also be destroyed.

f) An adequate plan for destroying the identifiers at the earliest opportunity, or justification for retaining identifiers.

No identifiers will be collected as part of the pre-screening process and no identifiers will be marked on the pre-screening documentation, which will be destroyed at the end of the pre-screening call after determining likely eligibility. Once recruitment to the study is complete (i.e. 200 subjects have successfully passed the screening process after giving informed consent) then the list of subjects who underwent the pre-screening process will also be destroyed. (See Section (e) above).

g) Written assurance that the PHI will not be re-used or disclosed for other purposes.

We provide assurance that the protected health information (PHI) will not be re-used or disclosed for other purposes except as required by law, or for authorized oversight of this research study.

h) Whether it is appropriate to provide the subjects with additional pertinent information after participation and, if so, how this will be accomplished.

As identifiers will not be collected during the pre-screening it is not appropriate for all the potential subjects pre-screened to be provided with additional information. Those subjects judged eligible to proceed on the basis of the phone pre-screening will be invited to a face-to-face meeting at the clinical site. There they will be provided with a copy of the Informed Consent Form, which the research coordinator will then talk through with them.

21.2 *Obtaining Informed Consent*

Informed consent will be taken by clinical trial coordinators at the clinical site. This will be done in a private room at the clinical site in a manner that is non-coercive and using language that is understandable to the

potential participants. No exculpatory language will be used, and the participants will not be asked to waive any of their legal rights. The subjects will be informed about the purpose and the duration of the current study, and the Informed Consent Form will be talked through with the subject. The procedures and objectives will be described. As will all investigations, potential discomforts, risks and benefits of participation. Sufficient time will be given to subjects to decide whether they wish to participate. Informed consent will be obtained, once the subject has been informed and wants to participate in this investigation, using the attached informed consent form.

In all cases of informed consent detailed below, the principal investigator (or approved designee) will sign the same consent form, on the same occasion as the subject. Three copies of the consent form will be made at UCSD and UU sites. The original copy of the signed consent documents will be kept in the Investigator site file at the investigation site. A copy will then be filed in the subject's medical notes at the clinical site, and a further copied provided to the subject. Where sites utilize electronic medical notes, a note of the patient's participation in this clinical investigation should also be made in the system. Two copies of the consent form will be made at the TDE site. The original copy of the signed consent documents will be kept in the Investigator site file at the investigation site and a scanned copy will be provided to the subject. Written informed consent from the subject must be obtained before any investigation related procedures (including screening procedures) are performed.

The investigator or another member of the investigative team should appropriately address all and any queries that a patient may have regarding the clinical investigation.

The investigator will return to the subject after sufficient time has elapsed and provide a recap of the clinical investigation. The approved consent form will then be provided to the subject in order to obtain written consent. In order that consent provided by the patient is voluntary and fully informed the subject should:

- Understand the purpose and nature of the investigation
- Understand what the investigation involves, its benefits (or lack of benefits), risks and burdens
- Understand the alternatives to taking part
- Be able to retain the information long enough to make an effective decision
- Be able to make a free choice and free form coercion
- Be capable of making this particular decision at the time it needs to be made.

21.3 *Special Circumstances for Informed Consent*

In accordance with the European Standard EN ISO 14155:2011, informed consent may be given by a legally authorized representative only if a patient is unable to make the decision to participate in the clinical investigation. In such cases, the subject will be informed about the clinical investigation within his/her ability to understand.

A consultee is a career or an individual who is concerned with the welfare of the named patient who lacks the capacity to give consent. The consultee will be consulted by the investigator as to whether the patient should take part in the clinical investigation and, in his/her opinion, what the patients' wishes would be in regard to participating if they had capacity in relation to the matter. The consultee will be enlisted to attempt explaining the key facts about the investigation in language that would likely be understood by the patient. Communication with the consultee may take place by face-to-face interview, telephone or correspondence by letter. Consultees will be provided with a detailed information sheet regarding the investigation and will be asked to sign a declaration form, 'Consultee Declaration Form' stating that the patient would not object to taking part in the clinical investigation.

For subjects or legally authorized representatives that are unable to read or write consent will be obtained through a supervised oral process. An independent witness shall be present throughout the process. The written informed consent form and any other information will be read aloud and explained to the prospective subject or his/her legal authorized representative and, wherever possible, either shall sign and personally date the informed consent form. The witness will also sign and personally dates the consent form attesting that the information was accurately explained, and the informed consent was freely given.

22. Subject Eligibility

Where a subject fails to fulfil the screening or randomization inclusion or exclusion criteria, this will be documented, and the investigator will retain the signed consent form. The subject will not advance any further into this clinical investigation.

23. Subject Identification

When a subject is considered eligible for entry into this clinical investigation, the subject will be allocated firstly an individual screening number, and should they meet the randomization criteria they will be allocated an individual number at randomization (subject ID number).

Subject number will consist of a unique numeral. These numbers will be unique identifiers of the subject and documented on each page of the Case Report Form or Requisition Form and all other documentation relating to the subject.

The Subject ID number will be assigned to participants at the UU site starting at U1001, at the UCSD CCR site starting at C1001, the UCSD EPARC site as E1001 and at the TDE site as T1001. They will then continue in a sequential order for each subsequent subject.

Logs relating to screening, enrolment and identification must be completed. However, the later log will only be held and the investigational site.

24. Treatment Procedure

24.1 *Randomization / Assignment of subjects to treatment groups*

Treatment assignments, investigation or sham devices will be generated via a randomized procedure, with participants randomly chosen in permuted blocks. At baseline, subjects will be assigned 1:1 between treatment (investigational device) and control groups (sham device). Stratification will be performed by gender and center.

24.2 *Blinding*

Externally the active and sham devices appear identical. They can only be distinguished by a unique serial number that is known only to the manufacturing team at Neurovalens. The master code will be emailed to the study statisticians, who have no direct subject contact. The statisticians developed an envelope based subject randomization system for the study coordinators, so as to maintain double blinding.

As per the FDA guidance the effectiveness of the blinding will be assessed at the 0 and 6 month timepoints. After using the device at the 1-month timepoint this will be via by a pop-up question in the study app asking the subjects to indicate which intervention group they believe they are in, and also why they believe this. The

coordinators will not be made privy to this information which will be passed to the study statisticians. The coordinators will be asked to provide the same information at 6 months (i.e. which group they believe the subject is in and why). At 6 months the coordinators will also ask the subjects to privately write down on a piece of paper what group they believe they are in and why. The coordinator should not look at the subject's answer until after the unblinding process has taken place by asking the subjects to indicate which intervention group they believe they are in, and also why they believe this. The coordinators will be asked to provide the same information at 6 months.

24.3 *Independent Data Monitoring Committee (DMC)*

There are no safety issues with the device that require a DMC for safety monitoring in this study.

24.4 *Prior to Initiating Study*

- The Sponsor and Investigator complete a Clinical Study Agreement
- The Investigator obtains Institutional Review Board (or other necessary regulatory/ ethical) approval of the protocol, any advertisements for the study, any information or other sheets to be given to participants and the Informed Consent Form.
- The Investigator signs the Investigator Agreement form.

24.5 *Pre-Testing Activities*

- Neurovalens will arrange delivery of the study materials required
- The study site staff will be trained on the IFU and how to connect the app on the iPod to the VeSTAL device. Training records shall be maintained in the Investigator Site File (ISF).
- The study site staff on this Clinical Study Protocol and any amendments. The training records shall be stored in the ISF.

24.6 *Hearing Examinations (At baseline and 6 months)*

The FDA provided feedback that study subjects should undergo a hearing test and otoscope examination of the ear canal. There is no suggestion from any of the literature looking at the safety of GVS that hearing, or the ear canal are adversely affected (Krystal et al., 2010; Marshall et al., 2010). And a study specifically looking at the impact of GVS on cochlear function showed no effect (Cevette et al., 2012). It is intended that an audiogram

(hearing) test be conducted on study participants at baseline and at 6 months. Prior to the audiogram, the outer canals of the study participant's ears will be examined with an otoscope to make sure that the tympanic membrane is intact, and that there is no infection. More details on this test are outlined below. Any change over the course of the study in a subject's hearing, or the condition of the external canal of their ear, will thus be determined.

Prior to the audiogram, the ear canals of the study participant will be examined with an otoscope to make sure they are free of wax, that the tympanic membrane is intact, and that there is no infection. This hearing test will be based on the standard and most common type of hearing test, pure tone audiometry. The result of the test is an audiogram diagram which plots a person's hearing sensitivity at the tested frequencies.

As described above the GSI AMTAS Flex system and video-otoscopy alternative may be deployed at the UCSD sites. This has been agreed with the FDA and will be carried out in a suitably quiet room. The PI at the UCSD sites, Dr Erik Viirre MD PhD, who is a clinical neuro-otologist will review the results of these assessments and decide if whether subsequent formal audiometry assessment is required.

25. Adverse Events

25.1 *General*

In accordance with the European Standard BS ES ISO 14155:2011 the following definitions apply in relation to adverse events.

Adverse Event (AE). Any untoward medical occurrence, unintended disease or injury, or untoward clinical signs (including abnormal laboratory findings) in subjects, users or other persons, whether related to the investigational medical device. For subjects this definition relates to all study procedures and comparators, for users and other persons it relates only to the investigational medical device.

Adverse Device Effect (ADE). Adverse event related to the use of an investigational medical device. This includes adverse events resulting from insufficient or inadequate instructions for use, deployment, implantation, installation, or operation, or any malfunction and events resulting from intentional misuse of the investigational medical device.

Serious Adverse Event (SAE). An adverse event that

a) Led to death,
b) Led to serious deterioration in the health of the subject, that either resulted in
    1) A life-threatening illness or injury, or
    2) A permanent impairment of a body structure or a body function, or
    3) In-patient or prolonged hospitalization, or
    4) Medical or surgical intervention to prevent life-threatening illness, injury or permanent impairment to a body structure or function.

Planned hospitalization for a pre-existing condition, or a procedure required by the CIP, without serious deterioration in health, is not considered a serious adverse event.

Serious Adverse Device Effect (SADE). Adverse device effect that resulted in any of the consequences characteristic of a serious adverse event.

Unanticipated Adverse Device Effect (UADE). This is defined by 21 CFR 812.3(s) as “any serious adverse effect on health or safety or any life-threatening problem or death caused by, or associated with, a device, if that effect, problem, or death was not previously identified in nature, severity, or degree of incidence in the investigational plan or application (including a supplementary plan or application), or any other unanticipated serious problem associated with a device that relates to the rights, safety, or welfare of subjects.”

26. Recording and Assessment of Adverse Events

All observed or volunteered adverse events (serious or non-serious) and abnormal laboratory findings, regardless of treatment group will be recorded in the subjects Case Report Form. For all adverse events, sufficient information will be obtained to permit 1) an adequate determination of the outcome of the event (i.e. whether the effect should be classified as a serious adverse event) and; 2) an assessment of the causal relationship between the adverse event and the investigational medical device or, if applicable, the other study treatment.

Adverse events or abnormal laboratory results felt to be associated with the investigational device or, if applicable, other study treatment or diagnostic products(s) will be followed until the event (or its sequelae) or the abnormal laboratory finding resolves or stabilizes at a level acceptable to the investigator-sponsor.

Abnormal laboratory results will be assessed by the Chief Investigator and in the event these results require additional follow-up, the subject will be contacted by the Chief Investigator (via email) advising the subject to contact their General Practitioner (GP).

In accordance with the standard operating procedures and policies of the local Independent Ethics Committee/ Institutional Review Board the site investigator will report SAE's to the IRB/IEC.

27. Causality and Severity Assessment of Adverse Events

The investigator-sponsor will promptly review documented adverse events and abnormal laboratory results to determine 1) if the abnormal laboratory finding should be classified as an adverse event; 2) if there is a reasonable possibility that the adverse event was caused by the investigational medical device or, if applicable, other study treatment or diagnostic product(s); and 3) if the adverse event meets the criteria for a serious adverse event.

If the final determination of causality is unknown and of questionable relationship to the investigation medical device or, if applicable, other study treatment or diagnostic products(s), the adverse event will be classified as associated with the use of the investigation medical device or, if applicable, other study treatment or diagnostic products(s) for reporting purposes. If the final determination of causality is unknown but not related to the investigational medical device or, if applicable, other study treatment or diagnostic products(s), this determination and the rationale for the determination will be documented in the respective subject's case report form.

28. Reporting UADEs

As per the FDA guidance in this matter (https://www.fda.gov/downloads/RegulatoryInformation/Guidances/UCM126572.pdf): UADEs must be reported by the clinical investigator to the sponsor and the reviewing IRB, as described below:

- For device studies, investigators are required to submit a report of a UADE to the reviewing IRB and the sponsor [Neurovalens] as soon as possible, but in no event later than 10 working days after the investigator first learns of the event (CFR 812.150(a)(1)).

• Sponsors must immediately conduct an evaluation of a UADE and must report the results of the evaluation to FDA, all reviewing IRBs/ equivalent competent authorities, and participating investigators within 10 working days after the sponsor first receives notice of the effect (CFR 812.46(b), 812.150(b)(1)).

29. Competent Authority Notification

It is the obligation and responsibility of the Sponsor to report to the Competent Authority all serious adverse events, serious adverse device effects and/or unexpected adverse device effects received from an investigator in a timely manner.

The sponsor will also inform all other participating principal investigators of all serious adverse events, serious adverse device effects and/or unexpected adverse device effects that are reported.

30. Follow-up of Unresolved Events

All serious adverse events, serious adverse device effects and/or unexpected adverse device effects will be followed up until they are resolved or for 30 days after the subject's participation in the clinical investigation ends.

31. Data Collection

All records generated as part of, or in relation to, this study shall be maintained or referenced in a study binder. These records will be maintained as required by applicable regulations and or guidelines after completion of the study.

Study site staff will be trained on the components of the study binder and the required documents which must be entered and documented in the study binder.

In response to a suggestion from the FDA that a site outside California be opened for the study, the sponsor opened a second site at the Ulster University in Northern Ireland, United Kingdom. This second site is at the Ulster University in Northern Ireland, at its Human Intervention Study Unit (See:

https://www.ulster.ac.uk/research/institutes/biomedical-sciences/core-facility-units/human-intervention-studies-unit). In order to increase the rate of rectuiment caused by the COVID19 pandemic, the TDE site were brought on board. Anonymized data from both all sites regarding the primary endpoints and safety endpoints will be sent to the statistical analysts. A poolability analysis will be carried out to assess the comparability of the non-US data and the US data.

The UCSD Office of Contract and Grant Administration have been asked to establish an unfunded collaboration agreement with the Ulster University All sites will act in accordance with GCP guidance and study monitoring will be in place. The Research Plan as executed at CCR UCSD will also be executed at the EPARC UCSD and Ulster University sites.

Data will be transferred in an encrypted PDF format. Trial staff will be given direction on how to share the trial data and given access to one specific section of a controlled cloud service which is controlled via role based access. Once they have authenticated with the service and the service verifies that they have the correct role to access the system they will be directed to a single webpage within the application where they will be able to upload the encrypted PDF. This PDF is generated on the fly and is therefore not stored in another location that could become compromised. Generating the PDF on the fly means that the source data is extracted from the database, processed and delivered in the context of a single request. This is an important factor as this means that after the request finishes the data in the PDF will not be held in application memory once the request is finished and the data relating to the request is freed. After the PDF is generated it is delivered to the assigned statistician using TLS 1.2 (a strong protocol), ECDHE_ECDSA with X25519 (a strong key exchange), and AES_128_GCM (a strong cipher). These data will be transferred for each subject when they complete participation in the study. If they complete the study protocol this will be at the six month mark, however, if they drop out early then data accrued up until that point will be sent.

31.1 *Completion of Case Report Form*

A Case Report Form may be in paper or an electronic format, details and procedures for completing Case Report Forms are provided in a separate document. All relevant investigation personnel are trained in the requirements of data collection and the method of collection prior to the first subject being recruited at that site. It is the responsibility of the PI to ensure that data is timely, accurate and complete.

31.2 *Device Usage Records During Home Use Period*

Within the system there are a number of stages that define how data gets from the VeSTAL wearable to the cloud service and subsequently the database:

1. Data logs are recorded by the VeSTAL wearable
2. The data logs are read from the wearable by the mobile application using Bluetooth Low Energy
3. The data logs are stored by the mobile application
4. When a full session is recorded the data logs for that session are sent to the cloud service using TLS
5. The cloud service parses the session data and stores it and the raw logs in the database

Along with this every record is date and time stamped at various points within the process. At point 1 a date/time stamp is generated on each log entry by the wearable, this is stored by the mobile application at point 3. At point 4 a start and end date/time stamp is generated for the entire session by the mobile application. At point 5 the session data is date/time stamped again to show when it was initially added to the cloud database. If any edits are performed after the data has reached the cloud service, the audit log will maintain a list of the following for each edit:

- A date/time stamp of when the change happened
- The user that made the change
- A snapshot of the data before the change
- A snapshot of the data after the change

31.3 *Review and Return of Completed Documentation*

The PI will make the original case report forms, where applicable, available to the monitor at each visit. Procedures for the return of completed documentation are provided in a separate document.

31.4 *Retention of Documentation*

The Principal Investigator, or designee, will retain all copies of the records for a period of 7 years from the completion of the clinical investigation. A list of essential documents to be maintained will be provided to each site at initiation.

- Clinical Investigation Plan (CIP - this document) and revisions
- Deviation Records

- Clinical site project-specific SOPs/protocols including any revisions
- Notes to File (NTF) (for clarifications only – not to be used for any deviation from this TSP)
- Financial disclosures
- Any Investigator Agreement(s)
- IRB/ethics approval: approval (including revisions), roster and annual reports/ final IRB report if applicable.
- Clinical site certification
- Training: Device-specific training and proficiency records
- Training records for all operators involved in receipt of the device, testing with the device and/or reporting of results, including review
- Equipment: maintenance records
- Records for device:
    - Device shipping records, receipt, use, disposal and/or return records
    - Records of exposure to device (test records)
    - Source records
- Clinical site monitoring records:
    - Monitoring log and monitoring attendance log supplied by Neurovalens at each monitoring visit
    - Site Initiation Visit report, monitor updates if applicable and memo from monitor to start testing
    - Routine monitoring visit reports
    - Site Close Out Visit report
- Device adverse events
- Correspondence relating to the device and this plan:
    - Key emails
    - Key conversation phone records

Note: It is likely that multiple binders/files will comprise the “regulatory binder”. Assessment of maintenance of the study binder will be conducted at test site monitoring visits.

32. Suspension, Termination and Close-Out of Clinical Investigation

32.1 *Suspension or Premature Termination of Clinical Investigation*

Reasons for suspension or premature termination at an investigation site may include incidences where monitoring or auditing identifies serious or repeated deviations on the part of an investigator. The FDA and IRB will be notified of any suspension or early termination of the clinical investigation, together with all other principal investigators, in the event that the suspension or termination was due to safety issues. A principal investigator may suspend or prematurely terminate participation in the clinical investigation at the investigation sites for which they are responsible.

If suspicion of an unacceptable risk to subjects arises during the clinical investigation, or when so instructed by the IRB or regulatory authorities, the clinical investigation will be suspended while the risk is assessed. The clinical investigation will be terminated if an unacceptable risk is confirmed. Should the risk not be confirmed justification and data supporting the decision to resume the clinical investigation will be provided to the relevant persons.

32.2 *Routine Study Termination and Close Out*

Routine close-out procedures will be conducted ensuring that the PI's records are complete, all documents needed for the sponsor files are retrieved, remaining clinical investigation materials are disposed of, previously identified issues have been resolved.

33. Data Management Procedures

The investigator will prepare and maintain adequate and accurate source documents designed to record all observations and other pertinent data for each enrolled subject. Study Staff at each site will enter data from source documents corresponding to a subject into the protocol- specific Case Report Form. Subjects shall not be identified by name in the study database or on any study documents but will be identified by a site subject number. The handling of all data on the Case Report Form will be the responsibility of the PI.

The de-identified data will be entered into a validated database. Once all the quality assurance steps have been completed and the data integrity verified the database shall be locked. Analysis of these anonymized data will be conducted according to a pre-determined statistical analysis plan.

Data from subjects who received treatment, who withdraw voluntarily or who are withdrawn from the clinical investigation, will be used in the final analysis. The inclusion of partial data will be documented in the final report.

All procedures for the handling and analysis of data will be conducted using good computing practices meeting FDA guidelines for the handling and analysis of data for clinical trials.

33.1 *Data Archiving*

The database is safeguarded against unauthorized access by established security procedures; appropriate backup copies of the database and related software files will be maintained. Databases are backed up by the database administrator in conjunction with any updates or changes to the database.
At critical junctures of the protocol (e.g., production of final reports), data for analysis is locked and cleaned per established procedures.

34. Statistical Plan

Prior to analysis of the final study data a detailed statistical analysis plan will be written describing all analyses that will be performed.

34.1 *Statistical Plan*

Analysis Populations

The Primary Analysis population will be the 200 subjects included in the study. The primary analysis population is defined to include all randomized subjects (i.e. an intention-to-treat population).

Representativeness Analysis

To evaluate the representativeness of the population the following analysis will be conducted on the:

Primary Analysis Population:

As per the guidance from the FDA relating to the representativeness of the study population (https://www.fda.gov/downloads/MedicalDevices/DeviceRegulationandGuidance/GuidanceDocuments/UCM507278.pdf.source=govdelivery&utm_medium=email&utm_source=govdelivery) the following analyses will be conducted:

For each demographic characteristic an appropriate statistical test will be conducted to flag any differences between the two respective subgroups (active and sham devices). For categorical data either a Chi-square test (if $n \geq 5$ in each table cell) or Fisher's exact test (if $n < 5$ in any single table cell), will be used, and for continuous a 2-sample t-test will be used to compare the respective subgroups.

Missing data will be clearly reported and included in tables, tests for representativeness will be conducted only on the data actually present.

Randomization

Treatment assignments, investigation or sham devices will be generated via a randomized procedure, with participants randomly chosen in the permuted blocks. At baseline subjects will be assigned 1:1 between the treatment (investigational device) and control groups (sham device). Stratification will be performed by gender and center.

Methods: Primary Aims

For total body weight loss (TBWL) as a continuous measure, we will use a linear regression with TBWL at each time period as the dependent variable and group and other covariates of interest (baseline total body weight, gender, age, study site) as independent variables. As a supportive analysis, we will also a two-sample t-test to examine the differences between the control and active treatment groups at each time period. To assess any longitudinal effects, we will use a linear mixed effects model for weight over time, with fixed covariates for baseline weight, time, group, and a time / group interaction, and a random intercept to account for within-subject correlation. We will use these models to generate two-sided 95% confidence intervals for the difference in mean percent weight loss.

We will calculate exact binomial confidence intervals (95%, two-sided) to determine if the response rate in the treatment group significantly exceeds 50%, with response defined by a loss of at least 5% of their body weight at the 6-month follow-up. As supportive analyses we will also use Fisher's exact test to compare the response rates in each group, and, will consider the use of a logistic regression to control for any other covariates of interest. To investigate possible longitudinal trends, we will fit a logistic mixed effects model, similar to the linear mixed effects model described above.

Methods: Secondary Aims

For each continuous and categorical variable at each time point, we will use a two-sample t-test and Fisher's exact test, respectively. Summary statistics (mean, standard deviation, quantiles, counts, and percentages) and plots will be produced for all demographic and study outcomes. We will investigate the effectiveness of the device by usage by including usage time as a continuous covariate in a linear model for total fat loss. Statistical personnel will conduct all analyses using the latest version of R (R Foundation for Statistical Computing, Vienna, Austria. http://www.R-project.org/).

We will analyze mean fat loss in the same way as TBWL as a continuous measure with t-tests, linear regression and linear mixed effects models.

Missing Data

The primary hypotheses will be tested on the intent-to-treat population. Missing data will be clearly reported and included in tables. We will assess the assumption that the data is missing completely at random. For the primary effectiveness analysis of total body weight loss (TBWL) as a continuous measure we will perform a multiple imputation analysis. We will include the same covariates in the multiple imputation model as in the linear regression, i.e. group, baseline total body weight, gender, age, study site. Two supportive sensitivity analyses with different approaches to imputing missing data will also be performed: last observation carried forward (LOCF) and baseline value carried forward (ie assuming no weight loss).
A sensitivity analysis will be performed with the binary weight loss endpoint. This analysis will include a best-case scenario (assuming subjects with missing values have all lost over 5% body weight), a worst-case scenario (assuming no subjects with missing values have lost over 5% body weight), and a tipping point analysis (as described in Yan et al., 2009).

Interim Analysis

There is no planned interim analysis.

34.2 *Primary Endpoints USA Regulatory Clearance:*

The efficacy of the VeSTAL device within the treatment group will be quantified by calculation of two primary endpoints after 6 months treatment:

- mean percent weight loss;
- proportion of participants who lose at least 5% body weight.

Study Acceptance Criteria USA:

- Mean: There should be at least a 2% total body weight loss (TBWL) superiority margin between the active-product and sham-treated groups.
- Categorical: The proportion of participants who lose 5% TBWL or more in the active-product group is at least 50%, independent of the sham control

Primary Outcome Measures USA (6 months)

- Mean: There should be at least a 2% total body weight loss (TBWL) superiority margin between the active-product and sham-treated groups.
- Categorical: The proportion of participants who lose 5% TBWL or more in the active-product group is at least 50%, independent of the sham control.

Both weight loss efficacy acceptance criteria must be met to claim success. These criteria can be specified in mathematical formulae as follows:

Rejection of both the null hypotheses $\mu A - \mu C \leq 2\%$ and $\pi A \leq 50\%$ at the 2.5% (1-sided) level

Where:

μA and μC are the underlying population mean percentage weight loss on active and control respectively

πA is the underlying population proportion of subjects who lose ≥5% of baseline body weight on active.

Secondary Endpoints USA

A variety of secondary endpoints will also be assessed statistically at the 6-month timepoint after the primary endpoints. The following of these may form the basis of potential future labelling claims:

- Total body fat loss (as measured by whole body DXA scanning);
- Healthy Eating Index (HEI) score (https://epi.grants.cancer.gov/hei/) (as measured by the two-day 24-hour dietary recall);
- Total caloric intake (as measured by the two-day 24-hour dietary recall);
- Systemic inflammation (as measured by high-sensitivity C-reactive protein);

- Atherogenic index (as determined by the ratio of total cholesterol to HDL);
- Quality of life (as measured by the IWQoL questionnaire).

The full details of the testing procedures procedure for these endpoints will be laid out in the Statistical Analysis Plan. All other secondary endpoints at the 6-month timepoint will be hypothesis generating only.

Secondary Outcome Measures USA (At 6 months)

- The difference in mean fat mass loss (in grams) between the active-product and sham-treated groups is statistically significant. (As measured by means of a whole body DXA scan).
- The difference in mean trunk fat loss (in grams) between the active-product and sham-treated groups is statistically significant. (As measured by means of a whole body DXA scan).
- The difference in mean visceral adipose tissue (VAT) mass loss (in grams) between the active-produce and sham-treated groups is statistically significant. (As measured by the whole body DXA scan).
- Difference in mean lean mass gain (in grams) between the active-product and sham-treated groups is statistically significant. (As measured by means of a whole body DXA scan).
- Difference in mean bone mineral content (BMC) gain (in grams) between the active-product and sham-treated groups is statistically significant. (As measured by means of a whole body DXA scan).
- Fasting glucose
- Lipid profile (Assess total non-HDL cholesterol, and HDL to total cholesterol ratio)
- High-sensitivity C-reactive protein
- Glycated hemoglobin (HbA1c)
- Two-day 24-hour dietary recall
- Modifiable activity questionnaire
- Blood pressure
- Heart rate
- Waist and hip circumferences (This will only be performed at the USA sites.)
- Body Mass Index (BMI)
- Quality of life – IWQOL-Lite questionnaire.

- Dose response analysis: it is likely that some subjects will not utilize their device for the mandated 7 hours per week. Usage data will be available from the devices permitting a dose response analysis, which will done be in an intention to treat manner.
- Adjustment of medication – both reduction and new medications.
- Mentor support group usage (total hours).

34.3 *Primary Endpoints EU Regulatory Clearance:*

The efficacy of the VeSTAL device within the treatment group will be quantified by calculation of one endpoint:

1. Mean percent weight loss

Study Acceptance Criterion EU:

1. Mean: There should be at least a 2% total body weight loss (TBWL) superiority margin between the active-product and sham-treated groups.

Primary Outcome Measure EU (At 6 months)

1. Mean: There should be at least a 2% total body weight loss (TBWL) superiority margin between the active-product and sham-treated groups.

This criterion can be specified in formulae as follows:

$mA - mC \geq 2\%$ and $pt \leq 0.05$

Where:

mA and mC are the observed mean percentage weight loss on active and control respectively

pt is the p-value (two-sided) from a linear regression comparing percentage weight loss between active and control ($H0: \mu A = \mu C$; $H1: \mu A \neq \mu C$)

μA and μC are the underlying population mean percentage weight loss on active and control respectively

Secondary endpoints which may become the basis of claims in EU:

A variety of secondary endpoints will also be assessed statistically at the 6-month timepoint after the primary endpoints. The following of these may form the basis of potential future labelling claims:

- Total body fat loss (as measured by whole body DXA scanning);

- Healthy Eating Index (HEI) score (https://epi.grants.cancer.gov/hei/) (as measured by the two-day 24-hour dietary recall);
- Total caloric intake (as measured by the two-day 24-hour dietary recall);
- Systemic inflammation (as measured by high-sensitivity C-reactive protein);
- Atherogenic index (as determined by the ratio of total cholesterol to HDL);
- Quality of life (as measured by the IWQoL questionnaire).

The full details of the testing procedures procedure for these endpoints will be laid out in the Statistical Analysis Plan. All other secondary endpoints at the 6-month timepoint will be hypothesis generating only.

Other Secondary Outcome Measures EU (At 6 months)

- The difference in mean fat mass loss (in grams) between the active-product and sham-treated groups is statistically significant. (As measured by means of a whole body DXA scan).
- The difference in mean trunk fat loss (in grams) between the active-product and sham-treated groups is statistically significant. (As measured by means of a whole body DXA scan).
- The difference in mean visceral adipose tissue (VAT) mass loss (in grams) between the active-produce and sham-treated groups is statistically significant. (As measured by the whole body DXA scan).
- Difference in mean lean mass gain (in grams) between the active-product and sham-treated groups is statistically significant. (As measured by means of a whole body DXA scan).
- Difference in mean bone mineral content (BMC) gain (in grams) between the active-product and sham-treated groups is statistically significant. (As measured by means of a whole body DXA scan).
- Fasting glucose
- Lipid profile (Assess total non-HDL cholesterol, and HDL to total cholesterol ratio)
- High-sensitivity C-reactive protein
- Glycated hemoglobin (HbA1c)
- Two-day 24-hour dietary recall
- Modifiable activity questionnaire
- Blood pressure

- Heart rate
- Waist and hip circumferences (This will only be performed at the USA sites.)
- Body Mass Index (BMI)
- Quality of life – IWQOL-Lite questionnaire.
- Dose response analysis: it is likely that some subjects will not utilize their device for the mandated 7 hours per week. Usage data will be available from the devices permitting a dose response analysis, which will done be in an intention to treat manner.
- Adjustment of medication – both reduction and new medications.
- Mentor support group usage (total hours).

Power Calculation

The statistical plan was developed in conjunction with the biostatistics and trial design optimization company Exploristics (https://exploristics.com). Multiple simulation scenarios (each with 1000 iterations) were conducted in their KerusCloud platform to determine an appropriate study size for this evaluation. KerusCloud is a cloud-based, virtual simulation environment for optimizing clinical study design and analysis.

The primary outcomes are mean TBWL from the start of treatment (as a percentage of baseline total body weight), and the proportion of subjects who lose at least 5% of their baseline total body weight. These outcomes will be assessed at the 6-month timepoint.

Power calculations are based on simulation with assumptions informed by pilot data and the Modius weight-loss device as follows:

90kg median baseline weight in both groups (log-normal distribution with CV=24%)

6.6% mean percent weight loss in the active group at 6 months (SD 3.9%),

3.1% mean percent weight loss in the control group at 6 months (SD 3.9%),

correlations between baseline weight and weight loss at 6 months of -0.4.

Under these assumptions, with a sample size of 180 participants randomized 1:1 active: placebo, there is a greater than 80% power to demonstrate both the mean and categorical weight loss acceptance criteria at the 6-month timepoint.

Under an assumption of no difference between active and control but the same assumptions around SDs and correlations there is ≤2% probability of a type-I error with this sample-size.

Anticipating a drop out rate of 10%, we will plan to recruit n = 200 total subjects overall in the study across both sites.

35. Analysis of clinical performance

A Statistical Analysis Plan (SAP) will be written and finalized prior to any lock of the study database. The SAP will give a detailed description of the summaries and analyses that will be performed. The SAP is being prepared by the specialist statistical consultancy company Exploristics.

36. Reports and Publication

36.1 *Interim Report*

There is no planned interim analysis.

36.2 *Final Report*

Following enrollment completion and data monitoring of the clinical investigation a final report will be compiled in accordance with applicable regulations, even if the clinical investigation is terminated prematurely. The coordinating investigator will review, approve and sign off the final report.

36.3 *Publications*

Details for the preparation and submission for publication manuscripts containing the study results shall be determined by mutual written agreement OR documented in each Investigator Agreement. The publication or presentation of any study results shall comply with all applicable privacy laws.

As agreed in the Clinical Research Agreement (negotiated by OCGA) between UCSD and Neurovalens, “University shall have the right to publish the results of the work conducted by University under this Agreement, to the extent such results do not contain Confidential Information of Company, provided Company has the opportunity to review and comment on any proposed manuscripts describing said work thirty (30) days prior to their submission for publication. The Parties agree that the research and clinical investigation under this Agreement is part of a multicenter study, no publication or presentation shall be submitted or given by the University or Investigator until the study has been completed at all participating centers, or twelve (12) months has passed since the termination of the Agreement or completion of the Project, whichever is earlier University

agrees to reasonably consider Company's comments prior to publication. However, if such proposed manuscript contains patentable information, University will, at its option, either delete the patentable information and publish immediately, or withhold publication for up to an additional sixty (60) days to allow for the filing of patent applications."

37. Ethical Considerations

37.1 *Ethics Committee Approval*

Prior to the initiation of this clinical investigation, the Principal Investigator must ensure that the relevant research ethics committee approval is in place for his/her site. The following documents, along with any specific country or locally required documents, must be submitted; Clinical Evaluation Plan, Patient Information Sheet, Patient Consent Form must be submitted for approval.

37.2 *Subject Confidentiality*

Confidentiality of data shall be observed by all parties involved at all times throughout the clinical investigation. All data will be secured against unauthorized access. All subject data that appears in reports and publications will be anonymized such that the privacy and confidentiality of each subject is maintained.

In order to fulfil the requirements of the PI to allow access to source data, including where required, medical notes and meet confidentiality requirements the PI will be required to obtain consent from each subject stating that they agree for their medical records to be accessed where relevant to the clinical investigation (this will form part of the consent process).

1. Subject's information will be kept strictly confidential to the extent provided by law. An alphanumeric code will be assigned to each subject, and this code will be used in place of a name when reporting data. The hardcopy corresponding subjects with codes will be kept in a locked data cabinet in the study site, along with all results of all screening inventories, assays, and questionnaires. The computer to be used for data collection and analysis will require a password for access, which only personnel directly involved in the study will have. Data in any format will only be available for those on the protocol. Information collected in the study will not be reused or disclosed for other purposes. Data collected in the study will be destroyed after seven years.

2. All aspects of the study will be explained to the subject in lay language, and the experimenter will make sure that the subject understands what will happen prior to his/her participation. The experimenters will inform the subject of the precautions that will be taken with regard to personal information collected during the course of the study. The study will be as short as possible. Participation is voluntary, and subjects will be informed at the beginning of the study that they may withdraw at any time, for any reason, with no negative repercussions. The research should in no way result in social stigmatization or any other long-term distress to the subjects.

3. Once successfully passing the screening process we will share the follow subject details by email with the team at We Slim Together (WST): the subject's email address so they can be contacted by WST to enroll them in their program; details of the subject's hypocaloric diet plan; the subject's initial weight; and an ID number so we can identify the subject when liaising with WST. The subject's consent will be taken as part of the ICF in order to share these details.

4. If a subject enters their weight into the study app then this information (attached to the ID number WST have for that subject and devoid of any subject identifiable data) will be accessible by the subject's mentor at WST. This will be done by granting the mentor password-controlled access to login to a database. No other information from the application, other than the weight that the subject chooses to enter, will be made available to the WST team. This is only being done to help the mentor properly support the subject in their weight loss journey. This feature is detailed explicitly in the ICF and it is made clear to the subject that entering their weight into the VeSTAL application is optional. However, the subject is asked in the ICF to engage with their WST mentor and so, if they do not want to put their weight into the VeSTAL application, then they are asked to tell their mentor what their current weight is when they speak to them. Weight values entered into the app will not be accessible by any of the study personnel at the clinical site. And nor will any of these weight values be collected or used for any purpose in the study other than as stated above.

5. Once a month WST will email the study coordinator with a register as to how often the subjects are attending their weekly check in sessions with their mentor. This information will be linked to the subjects' ID numbers and not to their names or any other identifiable information. The subjects are notified in the ICF that WST will send us this information and are asked to consent to it and to agree to try to attend at least 75% of the mentoring sessions as part of taking part in the study.

6. No other data will be collected by the study staff from, or sent by the study staff to, any other social media sites or platforms – e.g. Facebook.

7. In order to minimize the risk of coercing subjects to participate, no attempts to recruit subjects during undergraduate classes will be made.

8. Similarly, due to the risk of coercion the following will be excluded from participating: graduate students in Psychology; undergraduate students working as research assistants at any of the study sites associated universities.

38. Declaration of Helsinki

This clinical investigation will be conducted in accordance with the relevant articles of the Declaration of Helsinki as adopted by the 18th World Medical Assembly in 1964 and as revised in Tokyo (1975), Venice (1983), Hong Kong (1989), South Africa (1996), Edinburgh (2000), Washington (2002), Tokyo (2004) and Seoul (2008).

39. Indemnity

Sponsor recognizes its liability in law to compensate for any injury sustained by a subject participating in this clinical investigation in accordance with the guidelines of the Association of the British Healthcare Industry (ABHI) for any subjects entered into the clinical investigation in the United Kingdom.

40. Compensation

There will be no monetary expense to the subject for participation in the study. All study visits will take place at the clinical sites, either the UU site, the CCR and EPARC at UCSD or the TDE site, and parking and reasonable traveling costs will be reimbursed. Scheduling of the sessions will be made to avoid inconvenience. The VeNS devices used in the study will be provided to the subjects free of charge, and the same applies to access to dietetic advice.

Participants in the study will receive £/$200 for their complete participation in this study. At both sites if they terminate their involvement early, then they will receive £/$50 for their baseline visit and £/$75 each for their 3 month and 6 month visits that they have completed. If a participant ends their involvement before completing the three-month visit, they will be asked to return their device and in return receive £/$15. The compensation

schedule below outlines the payments to be made to the participant at the UCSD, TDE and UU sites. This compensation is payment for the participants' time and not any type of coercion to participate. If subjects are excluded during the Screen Visit, then they will receive travel/parking expenses but no additional compensation.

Participants at UU and UCSD, TDE sites who complete the study for the 6 months duration will be given the supplied iPod used during the study as a gift.

Compensation Schedule:

Visit Details	EPARC	CCR	UU	TDE
Visit 1 (0 months)	$50	$50	0	$50
Visit 2 (3 months)	$75	$75	0	$75
Visit 3 (6 months)	$75	$75	£200	$75

41. Regulatory Requirements

This investigational device study has been designated as a non-significant risk study and subject to the abbreviated requirements of the Investigational Device Exemption regulation in the US in accordance with 21 CFR part 812.

Appropriate risk assessment shall be completed prior to shipping investigational materials. All appropriate labelling shall be attached to this protocol and where required, applied to the investigational components of the investigational system.

41.1 *Regulations and Standards*

US CFR Regulation	Description
21 CFR 820	Quality System Regulation (GMP)
21 CFR 814	Premarket Approval of medical devices
21 CFR 54	Financial Disclosure by Clinical Investigators

21 CFR 809.10	Labeling
21 CFR 812	Investigational Device Exemptions
21 CFR 50	Protection of Human Subjects
21 CFR 56	Institutional Review Boards
21 CFR 58	Good Laboratory Practice for Nonclinical Laboratory Studies
ISO Standards	**Description**
ISO 13485	Quality Management System
ISO 14971	Risk Management
ISO 14155	Clinical investigation of medical devices for human subjects. Good clinical practice
ISO 15223	Medical Devices - Symbols to be used with medical device labels, labelling and information to be supplied

42. Good clinical practice compliance

This Clinical Investigation will be conducted in accordance with the European Standard BS EN ISO 14155:2011 Clinical investigation of medical devices for human subjects – Good Clinical Practice and Note for Guidance on Good Clinical Practice: CPMP/ICH/135/1995, January 1997 (where applicable to medical devices). Parts of the Code of Federal Regulation (CFR21) will also be applied where applicable.

42.1 *Clinical investigation Personnel and Responsibilities*

Prior to initiation of this clinical investigation, each Principal Investigator will approve this CIP by signing the signature page. This signature confirms that the clinical investigation will be performed in compliance with the CIP. In addition, each investigator (including sub-investigators) will provide Financial Disclosure as laid out in 21CFR part 54.

The funding support for this investigator-initiated protocol will be from a gift to the Center for Brain and Cognition from Neurovalens.

42.2 *Site Monitoring*

The monitor will be responsible for securing the compliance of the clinical investigators to the signed agreement, the clinical protocol, the requirements of European Standard BS EN ISO 14155:2011, ICH GCP,

Relevant sections US CFR Title 21 Parts 50,56, and 312 and ICH Guidelines for GCP (E6) or conditions of approval imposed by the reviewing competent authorities or ethics committee.

The principal investigator will permit a representative from or on behalf of Sponsor to inspect all Case Report Forms and corresponding portions of the subject's clinic records and/or original hospital medical records to ensure accuracy, at regular intervals throughout the clinical investigation. These inspections are for the purpose of verifying adherence to the protocol and the completeness and accuracy of the data being entered on the Case Report Forms.

The planned extent of source data verification is as follows:

- Subjects screened but not randomized will have initial inclusion and exclusion criteria verified;
- Randomized subjects will have all applicable data verified (20% Source Data Verification);
- All subject consent form will be verified.

An Investigator found not to comply will receive written notification of the deficiency, which will include a request that the deviation be corrected immediately. If the corrections are not made, shipments of products to the institution will be suspended and a request will be made to the Principal Investigator to return any products still in his/her possession to Sponsor.

42.3 *Auditing*

The relevant Regulatory Authority also has the right to conduct an audit of the clinical investigation, to determine that the investigation was, in fact, performed at stated investigation sites and that the data reported to the authority in support of a marketing application accurately reflects the data in the records of the clinical investigator. The authority also inspects such studies to verify that the clinical investigations were conducted in accordance with government regulations relating to the ethics committee and informed consent. It is the joint responsibility of the Sponsor and the Investigator to ensure that the clinical investigation has been completed in line with all government regulations for the relevant country.

In the event that the regulatory authority desires to inspect this clinical investigation, the investigator will permit authorized inspectors to inspect all facilities and records relating to the clinical investigation and aid the Inspector to perform the audit in a timely fashion.

42.4 *Modification to the Clinical Study Protocol*

Except in emergency situations, prior approval by the IRB is required for changes to, or deviations from, the protocol. In the event of such a situation, the PI must update the IRB expeditiously.

42.5 *Conflict of Interest*

Dr Ramachandran and Dr McGeoch are named as co-inventors on patent held by the Regents of the University of California that pertain to using electrical stimulation of the vestibular nerve as a means of altering body mass composition. Dr McGeoch was involved in co-founding Neurovalens, which has licensed the right to commercially exploit this technology from the University of California.

Neurovalens is funding the research as a grant via a Clinical Research Agreement negotiated with OCGA. Requisite forms have been filed for both Dr McGeoch and Dr Ramachandran with the CoI office at UCSD. The CoI forms have been reviewed by the IRC, which has outlined a CoI management strategy, which Drs Ramachandran and McGeoch have agreed to follow.

42.6 *Protocol Violations and Deviations*

A protocol violation or deviation occurs when the subject, investigator or Sponsor fails to adhere to protocol requirements affecting the inclusion, exclusion, subject safety, device testing, and any primary endpoint criteria. Any violation or deviation to this protocol must be communicated to Neurovalens within two working days. Deviations must be recorded on the form provided as part of the study documentation pack. Description of the deviation shall include all study participants who are affected by the deviation.

Deviation: Any change, divergence, or departure from the approved study design or procedures of a research protocol that is under the investigator's control and that has not been approved by the IRB, and does not affect the participant's safety, rights, or welfare and/or the completeness, accuracy and integrity of the study data. This term though sometimes used interchangeably with the term "violation," is (i) most often used when the variance is intended for the safety of one or more research participants or is an unintended change that is not considered as serious as a violation, (ii) is considered minor or administrative, and (iii) may involve no more than minimal risk to participants or others.

Noncompliance: Failure to comply with federal regulations, state laws, institutional policies, requirements or determinations of the IRB, and/or provisions of the approved research study. It is not considered noncompliance when there is a need to deviate from the approved protocol in order to protect the welfare of research participants.

Protocol Violation: Any deviation that may affect the subject's rights, safety, or welfare, and/or the completeness, accuracy and integrity of the study data. This term though sometimes used interchangeably with "deviation" is often considered a major, more serious, variance from an approved protocol than a deviation.

Serious Noncompliance: Failure to comply with federal regulations, state laws, institutional policies, requirements or determinations of the IRB, and/or provisions of the approved research study, where the occurrence involves substantive potential or actual increased risk to the safety, rights and welfare of research subjects.

Failure to comply with Good Clinical Practice guidelines will also result in a protocol violation.

The Principal Investigator will determine if a protocol violation will result in withdrawal of a subject or reporting to the site IRB/IEC.

A deviation is considered a minor or administrative divergence from approved design and procedures when the deviation does not affect the subject's rights, safety, or welfare, and/or the completeness, accuracy and integrity of the study data. If a deviation occurs and meets this definition, then the deviation should be reported to the IRB on the Deviation Summary Log and submitted at the time of continuing review. At the time of continuing review, the Deviation Log will be reviewed to determine if continuing noncompliance has occurred.

43. References

Balaban, C. D. and B. J. Yates (2004). Vestibuloautonomic Interactions: A Teleologic Perspective. The Vestibular System**:** 286-342.

Bamshad, M., et al. (1998). "Central nervous system origins of the sympathetic nervous system outflow to white adipose tissue." American Physiological Society **R291**.

Bamshad, M., et al. (1999). "CNS origins of the sympathetic nervous system outflow to brown adipose tissue." Am. J. Physiol. 276 (Regulatory Integrative Comp. Physiol**. 45): R1569–R1578**.

Belgardt, B. F., et al. (2009). "Hormone and glucose signalling in POMC and AgRP neurons." J Physiol **587**(Pt 22): 5305-5314.

Bent, L., R, et al. (2006). "Modulation of muscle sympathetic bursts by sinusoidal galvanic vestibular stimulation in human subjects." Exp Brain Res **174**: 701-771.

Bikson, M., et al. (2009). "Establishing safety limits for transcranial direct current stimulation." Clin Neurophysiol **120**(6): 1033-1034.

Bolton, P. S., et al. (2004). "Absence of short-term vestibular modulation of muscle sympathetic outflow, assessed by brief galvanic vestibular stimulation in awake human subjects." Exp Brain Res **154**(1): 39-43.

Brunoni, A. R., et al. (2011). "A systematic review on reporting and assessment of adverse effects associated with transcranial direct current stimulation." Int J Neuropsychopharmacol **14**(8): 1133-1145.

Brunoni, A. R., et al. (2012). "Clinical research with transcranial direct current stimulation (tDCS): challenges and future directions." Brain Stimul **5**(3): 175-195.

Brunoni, A. R., et al. (2013). "The sertraline vs. electrical current therapy for treating depression clinical study: results from a factorial, randomized, controlled trial." JAMA Psychiatry **70**(4): 383-391.

Carmona, S., et al. (2011). "Galvanic vestibular stimulation improves the results of vestibular rehabilitation." Ann N Y Acad Sci **1233**: E1-7.

Cone, R. D. (2005). "Anatomy and regulation of the central melanocortin system." Nat Neurosci **8**(5): 571-578.

Fitzpatrick, R. and B. Day, L (2004). "Probing the human vestibular system with galvanic stimulation." J Appl Physiol **96**: 2301-2316.

Fujimoto, C., et al. (2016). "Noisy galvanic vestibular stimulation induces a sustained improvement in body balance in elderly adults." Sci Rep **6**: 37575.

Fuller, P. M., et al. (2004). "Evidence for macular gravity receptor modulation of hypothalamic, limbic and autonomic nuclei." Neuroscience **129**(2): 461-471.

Fuller, P. M., et al. (2000). "Effects of 2-G exposure on temperature regulation, circadian rhythms, and adiposity in UCP2/3 transgenic mice." J Appl Physiol **89**: 1491–1498.

HaloNeuroscience (2016). "Safety of Non-Invasive Brain Stimulation delivered via the Halo Neurostimulation System in Healthy Human Subjects." from https://halo-website-static-assets.s3.amazonaws.com/whitepapers/safety.pdf.

Hao, Z., et al. (2016). "Reprogramming of defended body weight after Roux-En-Y gastric bypass surgery in diet-induced obese mice." Obesity (Silver Spring) **24**(3): 654-660.

Harris, R. B. S. (1990). "Role of set-point theory in regulation of body weight." The FASEB Journal **Vol 4**: 3310-3318.

Herrera, B. M., et al. (2011). "Genetics and epigenetics of obesity." Maturitas **69**(1): 41-49.

Horvath, T. L. (2005). "The hardship of obesity: a soft-wired hypothalamus." Nat Neurosci **8**(5): 561-565.

Jeong, J. K. and S. Diano (2014). "Prolyl carboxypeptidase mRNA expression in the mouse brain." Brain Res **1542**: 85-92.

Kessler, S. K., et al. (2012). "Differences in the experience of active and sham transcranial direct current stimulation." Brain Stimul **5**(2): 155-162.

Krystal, A., et al. (2010). "The Effect of Vestibular Stimulation in a FourHour Sleep Phase Advance Model of Transient Insomnia." Journal of Clinical Sleep Medicine **Vol 6** (No. 4).

Magis, D., et al. (2013). "Safety and patients' satisfaction of transcutaneous Supraorbital NeuroStimulation (tSNS) with the Cefaly® device in headache treatment: a survey of 2,313 headache sufferers in the general population." The Journal of Headache and Pain **14**(95).

Marshall, M., et al. (2010). "Therapeutic Effectiveness and Patient Acceptance of a Vestibular Nerve Activation Intervention in Chronic Insomnia." MEDICA MUNDI **54**(2).

McGeoch, P. D. (2010). The modulation of central pain by vestibular stimulation and another study on human brain function, Univeristy of Aberdeen.

McGeoch, P. D., et al. (2017).

McIntire, L. K., et al. (2014). "A comparison of the effects of transcranial direct current stimulation and caffeine on vigilance and cognitive performance during extended wakefulness." Brain Stimul **7**(4): 499-507.

Morales-Quezada, L., et al. (2015). "Cognitive effects and autonomic responses to transcranial pulsed current stimulation." Exp Brain Res **233**(3): 701-709.

Newlands, S. D., et al. (2003). "Central projections of the saccular and utricular nerves in macaques." J Comp Neurol **466**(1): 31-47.

Nitsche, M. A., et al. (2003). "Safety criteria for transcranial direct current stimulation (tDCS) in humans." Clinical Neurophysiology **114**(11): 2220-2222.

Paneri, B., et al. (2015). "The tolerability of transcranial electrical stimulation used across extended periods in a naturalistic context by healthy individuals." peer J Preprints **4**.

Parton, L. E., et al. (2007). "Glucose sensing by POMC neurons regulates glucose homeostasis and is impaired in obesity." Nature **449**(7159): 228-232.

Pialasse, J. P., et al. (2015). "The Vestibular-Evoked Postural Response of Adolescents with Idiopathic Scoliosis Is Altered." PLoS One **10**(11): e0143124.

Pialasse, J. P., et al. (2015). "Sensory reweighting is altered in adolescent patients with scoliosis: Evidence from a neuromechanical model." Gait Posture **42**(4): 558-563.

Poreisz, C., et al. (2007). "Safety aspects of transcranial direct current stimulation concerning healthy subjects and patients." Brain Res Bull **72**(4-6): 208-214.

Raimundo, R. J., et al. (2012). "Lack of clinically detectable acute changes on autonomic or thermoregulatory functions in healthy subjects after transcranial direct current stimulation (tDCS)." Brain Stimul **5**(3): 196-200.

Russo, R., et al. (2013). "Perception of comfort during active and sham transcranial direct current stimulation: a double blind study." Brain Stimul **6**(6): 946-951.

Scheele, C. and S. Nielsen (2017). "Metabolic regulation and the anti-obesity perspectives of human brown fat." Redox Biol **12**: 770-775.

Shang, D., et al. (2015). "Melanocortinergic circuits from medial vestibular nuclei to the kidney defined by transneuronal transport of pseudorabies virus." Int J Clin Exp Pathol **8(2)**: 1996-2000.

Sims, E. A. H., et al. (1968). "Experimental Obesity in Man: Cellular Character of the Adipose Tissue." The Journal of Clinical Investigation **50**: 1005-1011.

Tadini, L., et al. (2011). "Cognitive, mood, and electroencephalographic effects of noninvasive cortical stimulation with weak electrical currents." J ECT **27**(2): 134-140.

Utz, K. S., et al. (2011). "Minor adverse effects of galvanic vestibular stimulation in persons with stroke and healthy individuals." Brain Inj **25**(11): 1058-1069.

Varela, L. and T. L. Horvath (2012). "Leptin and insulin pathways in POMC and AgRP neurons that modulate energy balance and glucose homeostasis." EMBO Rep **13**(12): 1079-1086.

Vignaux, G., et al. (2013). "Bone remodeling is regulated by inner ear vestibular signals." J Bone Miner Res **28**(10): 2136-2144.

Wang, D., et al. (2015). "Whole-brain mapping of the direct inputs and axonal projections of POMC and AgRP neurons." Front Neuroanat **9**: 40.

Wilkinson, D., et al. (2009). "Safety of repeated sessions of galvanic vestibular stimulation following stroke: a single-case study." Brain Inj **23**(10): 841-845.

Yon, M. A., et al. (2013). "Relationships between dietary macronutrients and adult neurogenesis in the regulation of energy metabolism." Br J Nutr **109**(9): 1573-1589.

Zink, R., et al. (1998). "Effects of galvanic vestibular stimulation on otolithic and semicircular canal eye movements and perceived vertical." Electroencephalography and Clinical Neurophysiology **107**: 200–205.

Zink, R., et al. (1997). "Galvanic vestibular stimulation in humans: effects on otolith function in roll." Neuroscience Letters **232**: 171-174.

44. Appendices and Attachments

- **Safety Reporting -see below for safety reporting information and forms to assist in reporting requirements**
- **Investigational Device Instructions for Use**
- **Deviation/Violation Log**
- **EPIC-Norfolk Food Frequency Questionnaire (FFQ)**
- **Modifiable Activity Questionnaire (MAQ)**
- **Case Report Forms**
- **Example of Quality of Life Survey**

Safety Reporting

Safety reporting responsibilities to Sponsor as follows

	Who	When	How	To Whom
SAE	Investigator or Sponsor	Within 15 days of becoming aware of the event	SAE report form	Main IEC/REC for the investigation
Urgent Safety Measures	Investigator or Sponsor	Immediately	By Telephone, follow up written communication	Main IEC/REC for the investigation
Urgent Safety Measures	Exceptionally by local investigator	Within three days	In writing reason for the urgent safety measures and plan for further action/follow up	Main REC for the investigation