Oral **I**ron **R**epletion effects**ON** **O**xygen **U**p**T**ake in **H**eart **F**ailure: **IRONOUT-HF**

Cardiopulmonary Exercise Testing Manual of Operating Procedures

Massachusetts GeneralHospital Cardiopulmonary Exercise Laboratory

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1. Abbreviations and Glossary of Term

ABG	Arterial Blood Gas
AT	Anaerobic threshold
BP	Blood Pressure
CPET	Cardiopulmonary Exercise Test
CV	Coefficient of variation
ECG	Electrocardiogram
f	Breathing frequency
FEV_1	Forced vital capacity in 1 second
FVC	Force vital capacity
HF	Heart failure
HR	Heart rate
IC	Inspiratory capacity
MOP	Manual of Operating Procedures
MVV	Maximum voluntary ventilation (FEV_1 x 40)
O_2 Pulse	Oxygen uptake divided by HR
OUES	Oxygen Uptake Efficiency Slope
$PaCO_2$	Partial pressure of carbon dioxide
$P_{ET}CO_2$	Partial pressure of end tidal carbon dioxide
$P_{ET}O_2$	Partial pressure of end tidal oxygen
Peak VO_2	Peak oxygen uptake during exercise
PWC130	Physical Work Capacity at HR of 130 beats per minute
RER	Respiratory exchange ratio
SaO_2	Arterial oxygen saturation
SV	Stroke Volume
VCO_2	Carbon dioxide output
VC	Vital capacity
V_D	Dead Space Volume
V_E	Minute ventilation
VO_2	Oxygen uptake
V_T	Tidal volume
W	Watts

2. SITE ASSESSMENT AND CERTIFICATON

2.1 INTRODUCTION

The IRONOUT-HF Trial will utilize cardiopulmonary exercise testing (CPET) to derive peak VO_2 at baseline and after 16 weeks of treatment with oral iron repletion or placebo. Change in peak VO_2 will be the primary endpoint. CPET will also permit measurement of clinically relevant secondary endpoints such as workload achieved, anaerobic threshold and other indicators of cardiovascular reserve capacity.

Previousmulticenter trials evaluating exercise gas exchange have been confounded by methodological differences in exercise protocols and lack of uniformity in interpretation of gas exchange data.[1-3]The Heart Failure NetworkCPET Core Laboratory at Massachusetts General Hospital will work with individual centers to promote uniformity in CPET administration, reporting, and quality control measures. The CPET Core Laboratory recognizes that most participating laboratories have significant experience and expertise in administering CPETs. However, site assessment, certification, and strict adherence to this detailed protocol will be essential to ensure the consistency and validity of derived results. This manual has been divided into 8 sections, which include printable forms to guide sites through qualification procedures, patient education, and CPET administration.

2.2 CPET EQUIPMENT REQUIREMENTS

Cycle/Treadmill: Cycle ergometry will be the preferred exercise modality for IRONOUT-HF Trial. For CPET laboratories that do not perform cycle ergometry an alternative treadmill exercise protocol has been devised (Section 3). Regardless of the exercise modality, the metabolic cart computer should be able to control the work rate of the cycle or treadmill. Electrically-braked cycles will be required, as opposed to friction-braked cycles, based on their higher precision of work rate relative to friction-braked cycles and the ability to implement a continuous ramp protocol.[4] It will be essential to conduct the qualifying CPETs on the same equipment that will be used for the CPETs conducted for this trial.

Airflow or volume transducers: The accurate measurement of ventilation parameters during exercise is critically dependent on the accuracy of the flow-sensing device. Transducers used in exercise testing should meet established standards by the American Thoracic Society for flow and volume measurement during spirometry.[6]

Gas analyzers: Breath-by-breath analysis requires precise knowledge of gas analyzer delays and response kinetics.[7] Participating laboratories will need to follow standards for gas analyzer performance in breath-by-breath mode; these will include a transfer delay time of <1 second, a rise time <0.1 seconds, calibration stability of ± 3% over 20 min and calibration linearity ± 3% over the entire range.[8] Each site will be required to maintain a calibration logbook so that long-term trends can be monitored.

Electrocardiographic monitoring: Participating laboratories will be required to use electrodes and detection electronics designed for movement artifact rejection. Silver or silver chloride electrocardiogram (ECG) electrodes with circumferential adhesive provide good electrical

contact and minimize movement artifact. Continuous display of ECG tracings with 12-lead ECG placement as described by Mason and Likar.[10] The timing of ECG monitoring must be synchronized with the timing used by the gas exchange system, preferably through an integrated ECG-metabolic cart system. ECGs will be performed for precise heart rate monitoring and for safety purposes, but will not be transmitted to the core lab for interpretation.

Metabolic measurement systems: The core laboratory encourages sites to utilize standard metabolic cart processing software.This will promote uniform generation, formatting and acquisition of breath-by-breath data. Medgraphics Inc (St. Paul, MN) metabolic carts interfaced with BREEZESUITE software represent the most commonly used metabolic measurement systems in the United States and the primary equipment used by the Core Laboratory. Therefore CPET data acquired and configured with BREEZESUITE software is preferred. The second most common type of metabolic cart used in CPET is Viasys (previously Sensormedics), which the core lab is equipped to interpret. For Viasys/Sensormedics equipment, sites will be strongly encouraged to use ENCORE/Vmax software formatted data to facilitate data manipulation and simple transfer of data to the core laboratory. If an alternative metabolic measurement system is utilized (i.e. Parvomedics), it must allow real-time tabular and graphical display of exercise variables, 5-of-7 breath moving average integration of gas exchange variables, and data conversion to unencrypted format such as Excel that will lend itself to interpretation by the core laboratory.

2.3 CPET EQUIPMENT CALIBRATION

CPET equipment should be calibrated by following instructions given by the manufacturer of the equipment, and in accordance with the schedule outlined below. Participating sites will be required to maintain a calibration logbook.

Prior to performance of CPETs for IRONOUT-HF,the following calibration procedures are recommended but not required.

1. Electrically braked cycle ergometers that have not been previously calibrated or newly purchased should be dynamically calibrated with the use of a dynamometer (torque meter). Because many labs do no have dynamometers, cycle ergometer manufacturers may be required to provide this service. This calibration should be repeated if the cycle is moved or jarred or if certification testing results in abnormal values for gas exchange-work rate relationships.
2. Treadmills should have belt speed verified by timing revolutions using a mark made on the treadmill belt with a subject on the treadmill. Grade may be determined by using a plumb line and tape measure.

Every 12 months

1. Physiologic calibration: A healthy volunteer, consuming a stable diet performs a constant work rate test (i.e. 50 W and 100 W) in which steady state values for V_E, VCO_2, VO_2 are compared to those of previous tests. Coefficients of variation (%CV) should be below 5% for repeat oxygen uptake measurements and below 7% between repeat sessions, with values outside of the 95% confidence interval engendering system-wide reassessment.

Prior to each test

1. Record barometric pressure, temperature, relative humidity
2. Perform flow calibration with a 3L syringe (<1-15sec duration) to achieve ± 3% agreement with calculated volumes.
3. Perform gas analyzer calibration with two precision-analyzed gas mixtures. This is commonly done with one 6% CO_2 and 15% O_2 tank and one 0% CO_2 and 21% O_2 tank. The air baseline setting for O_2 and CO_2 should be checked before each test to correct for baseline drift since calibration.
4. Determine transport delays between the gas sampling point and each gas analyzer. This should be an automated process.

2.4 SITE QUALIFICATION PROCEDURES

Before baseline studies may be performed in subjects, each site will be required to submit two incremental symptom-limited CPET tests on a "standard normal subject". The standard subject should be a healthy adult. Sites have often used laboratory employees for qualification testing. These tests must be performed on separate days, preferably no more than 5 days apart, according to the protocol summarized in Section 7. Prospective CPET laboratories will be evaluated based on their ability to: (1) follow a site qualification protocol (see Figure 2 and Tables 7 and 8), (2) generate reproducible CPET data, and (3) transmit data to the core laboratory. Test results will be compared to data available on normal individuals from the core laboratory and the published literature.[8, 11] Sites should await feedback from the core laboratory confirming that their site has qualified prior to scheduling study patients for testing.

Sites may subsequently use repeated studies of the "standard normal subject" to verify accuracy of their systems. The Core Laboratory will require sites to maintain a detailed log of physiologic calibration testing as described above, but this information will not need to be transmitted to the core laboratory as part of the initial qualifying procedures.

In anticipation of transmitting qualification tests to the core laboratory the individual who will be transmitting data from the participating CPET laboratory to the core laboratory should take the following steps (also see Appendix 1): Email Diane Cocca-Spofford RN at dcoccaspofford@partners.organd CC glewis@partners.organd indicate the following:

   a. Regional research site name.
   b. Responsible CPET lab staff member who will primarily interact with the Core Laboratory with email address and contact information.
   c. Exercise modality that will be used for the clinical trials (cycle or treadmill)
   d. Metabolic cart manufacturer and software program that will be used.
2. Await receipt of an email from Diane Cocca-Spofford with an invitation to join Partners Research Computing network through which qualification study files and subsequent study files can be transferred via email (see Section 4 for detailed instructions).
3. Transmit the two qualification studies formatted as described below and summarized in Tables 7 and 8.

3.CPET PROCEDURES

3.1 CPET Preparation

The protocol for symptom-limited CPET testing forIRONOUT is displayed schematically in Figures 2 and 3. Cycle ergometry will be the preferred exercise modality with a ramped workload of10 watts/min For CPET laboratories that do not perform cycle ergometry an alternative treadmill exercise protocol has been devised that simulates cycle ergometry in terms of a linear, comparable increment in external work performed and a gradual increment in both speed and ramp that is appropriately suited to the study of HF patients.[12] In order to achieve within subject consistency, it is important that a laboratory that elects to perform treadmill ergometry commit to doing so for all study tests that it performs. In addition, if a participating laboratory has more than one ergometer or metabolic cart, the same equipment should be used for each test in a given subject.

1. **Patients should be given pre-test instructions according to your institution's CPET laboratory practice.** The Patient Education Form, Section 6 is provided as an example instruction sheet that can be used at the discretion of participating laboratories. Subjects should be given instructions to follow the same regimen of fasting (for at least 3 hours) and taking their medications prior to testing, particularly medications that influence heart rate.

2. **Review of contraindications to exercise testing**. Table 1 lists contraindications to exercise testing which are anticipated to be very rare within patients enrolled in IRONOUT-HF. Questions that arise regarding exercise eligibility should be brought to the attention of the site principal investigator.

Table 1: Contraindications to Exercise Testing:

Absolute Contraindications
• Acute myocardial infarction (3-5 days) or unstable angina • Uncontrolled symptomatic arrhythmias • Active endocarditis • Acute myocarditis or pericarditis • Symptomatic severe aortic stenosis • Acute pulmonary embolism or DVT • Suspected dissecting aneurysm • Uncontrolled asthma • Uncontrolled pulmonary edema • Room air desaturation to <85% • Acute non-cardiopulmonary disorder that may affect exercise performance (infection, orthopedic problem) • Mental impairment leading to inability to cooperate • History of exercise-induced ventricular arrhythmia

3. **Spirometric evaluation** should be completed upon arrival to the CPET laboratory by all sites that routinely include this test as part of the standard CPET procedure. This will

include forced vital capacity (FVC) and forced expiratory volume in 1 second (FEV1) from which maximum voluntary ventilation (MVV) will be estimated as (FEV1 x 35). There is not a requirement to perform spirometry at sites that perform spirometry separately from cardiopulmonary exercise testing.**Initial patient data entry:** Upon initiating a study using BREEZESUITE or analogous software, click on the "Patient" or equivalent demographic information tab. The following information should be entered into the designated text boxes.

Patient

Subject Identification: Enter study protocol, site number, subject number and initials, and the test description, with the designation A=baseline, B=16 week
IRONOUT_301_001_GDL_A
-For qualification studies, substitute the word Qualification for the protocol name and designate the study either A or B **(**i.e. **Qualfication_300_000_A)**.

Other demographics:Enter subject's date of birth, race, and sex

Visit Demographics

Enter the following:
Height (inches)
Weight (lbs)
Referring physician (CPET lab physician)
Technician (CPET lab technician)
Site: Indicate which site and metabolic cart are being used, if your laboratory has more than one

Patient History

Pre-test comments: Free text the name and contact information for the responsible CPET lab personnel who conducted the CPET
Name: _____Email: _____Phone #: _____

Free text spirometry data (if this data is not collected with an integrated program that automatically populates your metabolic cart software).
Forced expiratory volume in 1 second (FEV1)_____
Vital capacity (FVC)_____

4. **Record cycle seat height**. Before starting the test, the subject will sit on the cycle ergometer and CPET lab staff will adjust the seat height so that the subject's legs are almost maximally extended. Confirm this by having the patient turn the unloaded pedals several times so that he/she can become familiar with the cycle. Measure the seat height

for the patient with a ruler, and record it by entering free text into the "Patient History" section described above, so that the same height can be used for each subsequent study.

3.2 CPET procedures

3.2A. Blood pressure measurement procedures: Pre-exercise blood pressure should be obtained with the subject in a relaxed, comfortable seated position without clothing in between the cuff and the arm. Choose the correct cuff size, the bladder width should encircle 40% of the circumference of the arm and there should be at least 2 cm between the bottom of the cuff and the brachial artery. Record the blood pressure at which you hear the first Kortakoff sounds for two consecutive beats as systolic blood pressure and the pressure at which time the sounds disappear (K5) as diastolic blood pressure. CPET laboratory staff should adhere to this protocol in measuring exercise blood pressure as well.

3.2B. 12-lead electrocardiogram recording: Skin preparation is important to ensure a high-fidelity signal, free from motion artifact and electrical interference, that is sent to the electrocardiograph and the metabolic cart system where heart rate will be recorded. Proper skin preparation involves removing the hair with a disposable safety razor, cleansing the skin with alcohol or acetone to remove skin oils, followed by light abrasion to remove stratum corneum. Standard limb lead placement on the wrists and ankles must be modified for exercise by moving them to the anterior trunk in a Mason-Likar configuration (Fig. 1). A 12-lead electrocardiogram should be recorded during this rest period. Heart rate at rest and during exercise will be transmitted to the core laboratory. However, interpretation of cardiac rhythms and pattern interpretation on electrocardiograms will need to be performed contemporaneously with exercise testing (and not by the core laboratory) to ensure patient safety.

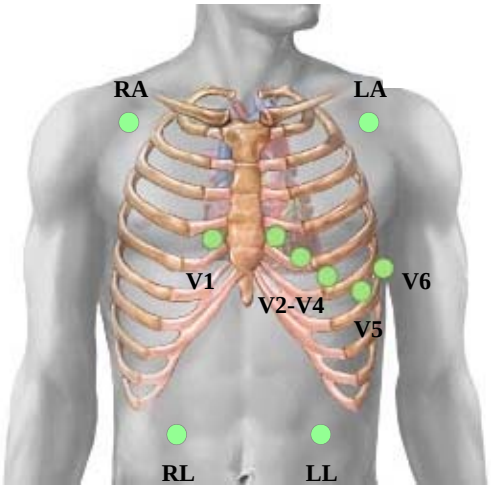


Figure 1. Electrode placement for electrocardiogram recording during exercise. RA indicates right arm, LA indicates left arm, RL indicates right leg, LL indicates left leg. Model adapted from ADAM at www.nlm.nih.gov/.../ency/imagepages/19865.htm

3.2C. Gas exchange measurements: With subjects sitting on the cycle ergometer or standing on the treadmill, a nose clip is placed and the mouthpiece is inserted. At this time the importance of maintaining a tight seal around the mouthpiece should be emphasized to the patient.

3.2D. Metabolic cart, software interface preset data displays: Metabolic measurement systems should allow real-time graphical and tabular display according to the recommended format in Table 4. Because significant differences can arise as a result of time interval selection for averaging breaths in patients with HF,[9] participating laboratories will be required to provide the core laboratory with data on all breaths. However, to standardize data output across metabolic carts we will request that tabular data be displayed using **5-out-of-7 breath retrograde time averaging** which is a standard option for BREEZESUITE, Viasys, and Jaeger Oxycon Pro metabolic cart software. The following screening procedures for outlier values should be programmed into Breeze Suite or analogous software systems.

Table 2. Parameters to eliminate outlier values for breath-by-breath gas exchange variables

Measurement Variable	Minimum Value
RER	0.5
VCO_2	50 ml/min
VO_2	50 ml/min
V_T	180 ml

RER indicates respiratory exchange ratio, VCO_2 carbon dioxide output, VO_2 oxygen uptake, and V_T tidal volume

Resting phase of CPET

A 5-minute rest period will be implemented for both protocols. During this period CPET lab personnel should observe key variables in comparison to reference values to ensure proper calibration and performance of the metabolic measuring system. **Table 3** provides an example of key resting variables and their expected values.[14]

Table 3. Reference values appropriate for resting conditions.

Heart Rate min^{-1}	VO_2 (ml/kg/min)	VO_2 (ml/min)	VCO_2 (ml/min)	RER	RR min^{-1}	V_E L/min	$P_{ET}O_2$ (mmHg)	$P_{ET}CO_2$ (mmHg)
60-100	3.5	200-300	140-300	0.7-1.0	12-20	6-10	100-105	38-42

VO_2 indicates oxygen uptake, VCO_2 indicates carbon dioxide output, RER respiratory exchange ratio, RR respiratory rate, V_E minute ventilation, $P_{ET}O_2$ end tidal oxygen, $P_{ET}CO_2$ end tidal carbon dioxide.

Some clinical conditions can account for departures from expected values. However, departures can usually be explained by pretest anxiety, leaks in the patient interface such as a poor fitting mask or failure to apply the nose clip or improper calibration of the metabolic cart. Anxiety is typified by a heart rate > 80 min^{-1},V_E> 10 L/min, $P_{ET}CO_2$ < 35,RER > 1.0 whereas a system leak results in proportionately lowV_E VO_2,VCO_2.

Warm up phase, unloaded cycling or walking on the treadmill: Following the rest period, at a verbal signal the patient should start pedaling with the cycle unloaded (free wheeling) for 3 minutes according to the appropriate protocol outlined in Table 9. Subjects undergoing treadmill testing who weigh <80 kg will follow Table 10a, subjects weighing >80kg will follow Table 10b. If available, an accessory motor should be utilized to rotate the flywheel at a rate of 60 rpm in order to eliminate the inertial force needed to start the flywheel rotating and reach the desired

speed. The patient should be coached to pedal at 60 rpm on the unloaded cycle to become accommodated to this pace. The pedal rate meter should be displayed in clear view of the patient to facilitate compliance with this goal pedaling frequency.

Incremental exercise: After 3 minutes of unloaded cycling, increase the work rate by 10 W/min using a continuous ramp protocol under computer control (see Table 9 for cycle ergometry and Table 10 for treadmill ergometry). Subjects will be encouraged to reach a maximal effort by monitoring the respiratory exchange ratio [goal respiratory exchange ratio (RER, VCO_2/VO_2) > 1.1] and by encouraging continued exercise until perceived exertion/dyspnea reaches a >8 on the Borg 0-10 scale.[15] We suggest printing out the Borg scale in large font so that patients can point to the scale during exercise. The technician and physician should work cooperatively to observe the patient's facial expression while encouraging the patient to keep their eyes open during exercise. Blood pressure, oxygen saturation, perceived exertion and dyspnea will be monitored and recorded every two minutes. ECG monitoring will be used to monitor patients during exercise testing. Heart rate, as measured by ECG, will be recorded every minute. The recognition and treatment of conditions that manifest with ECG abnormalities during exercise will be the responsibility of the on-site supervising physician due to the time-sensitive nature of such findings. If ECG abnormalities arise during testing these should be indicated in the commentary report section (see Section 7). Standardized guidelines for operators to stop an exercise test are listed in Table 4.

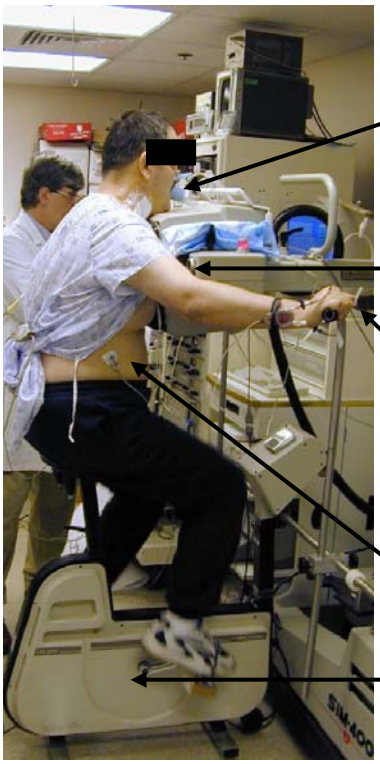
Criteria for CPET Termination: The most accepted criteria for terminating an exercise test are listed in Table 4. These are only guidelines and should be used in conjunction with clinical judgment of trained CPET lab personnel and study investigators.

Table 4. Objective criteria for termination of CPET:

Criteria for CPET Termination
Definitive ischemic ECG changes with associated chest pain
Complex ectopy (i.e. ventricular tachycardia)
Mobitz 2 Second Degree or Third degree heart block
Symptomatic fall in systolic blood pressure > 20 mmHg from the highest value during the test
Marked hypertension (systolic BP > 240 mmHg, diastolic BP >120 mmHg
Severe desaturation, SpO2<80% when accompanied by signs of severe hypoxia
Neurologic compromise such as mental confusion or loss of coordination

Recovery period: To avoid orthostatic hypotension when stopping exercise, the subject should be encouraged to slowly turn the pedals at 30 rpm to maintain venous return during the first 60 seconds of recovery. Record blood pressure during the second and fourth minute of recovery. Gas exchange and heart rate should continue to be measured for three minutes into recovery.

End of test: CPET laboratory personnel should elicit the reason for cessation of exercise. Viasys Vmax/Encore software prompts users with a "Metabolic End of Test Comments" menu. A primary reason for test cessation should be selected. Secondary reasons for cessation of exercise may also be recorded as added text. It is particularly pertinent to indicate if the test was stopped by the operator prior to the patient reaching a point of maximum exertion.



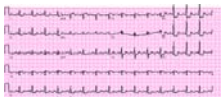
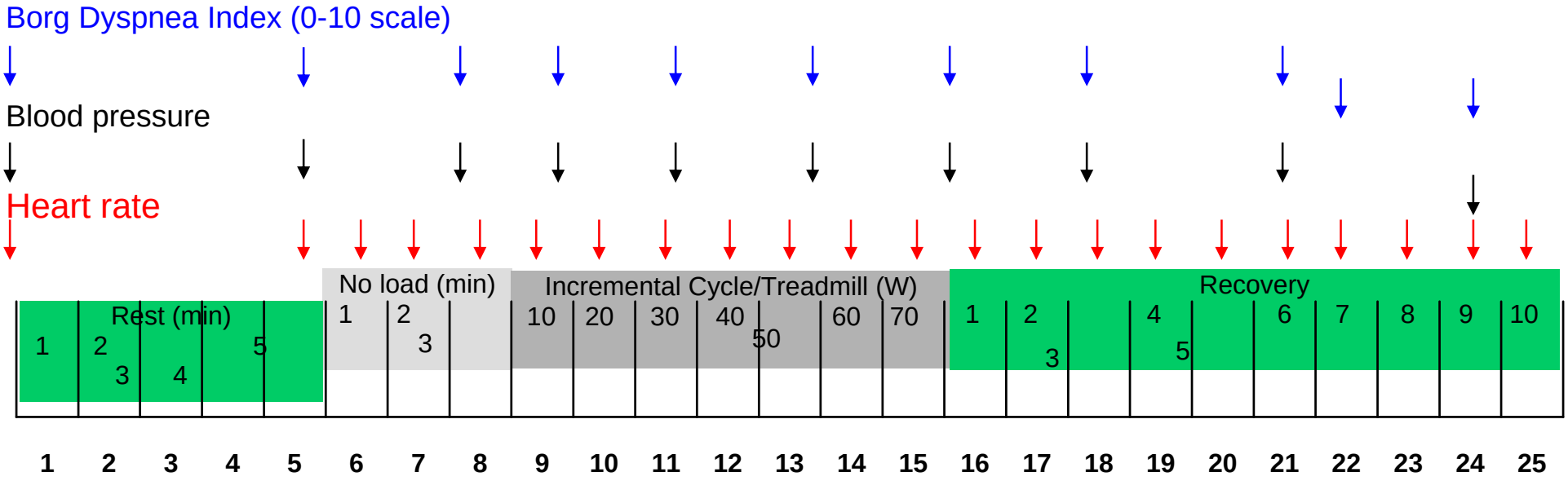
DEVICE	PRIMARY	DERIVED
Metabolic cart		
CO_2 O_2 Flow	$PETCO_2$, VCO_2 $PETO_2$, VO_2 RR, V_T, V_E	V_E/VCO_2 RER, AT O_2 pulse
	Blood pressure	Double product
Pulse Oximeter	SpO2	~O_2 saturation
	Heart rate Rhythm	Pattern changes
Cycle Ergometer 20 W/min ramp	Work Rate	Work efficiency

Figure 2. Measuring devices, primary, and derived measurements obtained during CPET. ET indicates end tidal, *f* breathing frequency, RER respiratory exchange ratio.



3.3 CPET Protocol Schematic Synopsis

Figure 3. Schematic representation of exercise protocols. Initial measurements of heart rate, blood pressure, and Borg Dyspnea Index will be conducted in subjects seated on the cycle ergometer prior to mouthpiece insertion. The 5 minute rest period will start upon mouthpiece insertion and initiation of gas exchange collection with the metabolic cart. Borg Dyspnea Index (Table 6B) and blood pressure will subsequently be obtained 4 minutes into the rest period, then during the last 30 seconds of each 2 minute increment during exercise (i.e. between 1.5 and 2.0 minutes into unloaded cycling, between 3.5 and 4.0 minutes into exercise etc). Peak exercise heart rate and Borg Dyspnea index should also be recorded.

3.4 Tabular CPET Data Display

Table 5. Tabular data displays for cardiopulmonary exercise testing.

Time (min:s)	Work (W)	VO_2 (ml/kg/ min)	VO_2 (ml/ min)	VCO_2 (ml/ min)	RER	RR (min^{-1})	Vt (ml)	V_E (L/ min)	$P_{ET}CO_2$ (mm Hg)	$P_{ET}O_2$ (mm Hg)	Cycle Rate (rmp)	HR (min^{-1})	O_2 sat (%)	SBP (mm Hg)	DBP (mm Hg)	Borg Dyspnea (0-10)

Unshaded cells represent standard metabolic cart output. The shaded regions of this table may be automatically populated during the study or may need to have data entered depending on the degree to which other measurement equipment is interfaced with the metabolic cart. To aid in data recording for post-test completion of these data fields, please see printable data entry forms (Tables 7-10). This information will need to be entered into the above tabular format either during or after the CPET, prior to transmission of the study to the core laboratory.

4. DATA RECORDING AND TRANSMISSION TO THE CORE LABORATORY

4.1 CPET Data Preparation: For each CPET performed the Core Laboratory requests that all information be integrated into a single CPET file that will be transmitted to the Core Lab. For each study the following steps will be taken to ensure standardized data reporting.

1. Prior to exercise testing, enter patient data into BREEZESUITE or analogous software according to instructions outlined above (Section 3.1, #5).

2. Data recording during CPET testing

A. Breath-by-breath data should be formatted according to Table 5.

B. Real-time addition of heart rate every minute, and blood pressure recordings and Borg scores every two minutes during exercise is technically possible in the BREEZESUITE and Vmax/Encore programs but may be cumbersome during supervision of a CPET. Hence, we recommend recording of heart rates (from the ECG), blood pressures and Borg scores on printed versions of Tables 9 or 10 (as appropriate) during testing. Following completion of the CPET, this data can then be entered into the Breeze Suite "Event Entry" screen or equivalent on other software systems. To access the Event Entry screen follow these steps

1. Enter the "Protocol/Log" tab, "Visit Log" section
2. Select the clock/pencil icon to enter data at an appropriate time during the test (i.e. 10 minutes of exercise)
3. Select the "Gas Exchange" tab with the "Event Entry" box
4. Use the drop down variable menu to select/enter Borg Score, BP, and heart rate
5. Select "ABG" to enter current hemoglobin level
6. Confirm that the Tabular data reporting form (Table 5) is updated for heart rate, blood pressure and Borg score recordings.
7. Add "Why the patient stopped exercising". A single primary reason should be specified, though addition of a secondary reason is permissible.

C. The core lab would prefer a self-contained single file in which all data is incorporated within the CPET software program. However, if this is not possible, data may be manually entered into electronic versions of Tables 8-10 and then transmitted to the core laboratory as a Microsoft Excel file. Files should be named by Protocol, site number, subject number, initials, and test description (baseline test will be A, second test B etc, i.e. IRONOUT**_300_001_GDL_A**).

4.2 CPET Data Transfer to the Core Lab: Copy the archived file onto a computer disk immediately following the CPET. BREEZESUITE software requires entering the Import/Export Program from the Start Menu→Programs→Medgraphics→DBP Tools→Tools→Export, then select the patient file and the destination of the file under the Brows menu. This will enable labs to export the file to a local disk and to then send the file electronically as an enclosure (see below). Viasys Vmax/Encore software can be used to directly email test files using the "Special Functions" tab and selecting the "File compress/email" option. Metabolic measurement systems with software programs other that those supported by Medgraphics and Viasys/Sensormedics will be required to convert gas exchange data into Excel format for breath-by-breath data

interpretation. In addition, graphical data should be generated to facilitate calculation of anaerobic threshold by the V-slope technique. Label the computer disk with the following information: a) HFCRN study name and Site name/ID#; b) patient ID and date of birth c) date of study. This will serve as a back-up hard copy of the data to remain at the individual CPET labs. Tests will only be mailed to the core lab upon request if there is a problem with electronic submission or archiving of the data. In the event that the core lab requests disks, the participating CPET lab should retain a copy of the data in their laboratory and send a disc via Fed Ex to:

Dr. Gregory Lewis
GRB 800, Cardiology Division
Massachusetts General Hospital
55 Fruit St, Boston, MA 02114

The MGH Core Laboratory will utilize the Accellion Secure File Transfer Service to exchange files with participating CPET laboratories via a web browser rather than ftp site. The service is a secure web based application with anti-virus detection built in. Attached files are encrypted and uploaded to an appliance and the recipient receives an email with a link. When the recipient clicks the link, the file is downloaded from the appliance. This will enable rapid, readily traceable transfer of data between participating sites and the core laboratory.

CPET laboratory personnel at participating sites will receive an invitation email from Diane Cocca-Spofford with instructions on how to register for this service. Upon clicking on the attachment the screen depicted in Figure 4 will appear with instructions on how to register for this free service. Participants will automatically create a login and password as part of a simple authentication process which can be used to send files back to the core lab or anyone else who is registered user with an affiliate email address. Participating CPET labs will be able to send up to 10 files at a time to the core laboratory, with an overall size limit of 2GB.

Figure 4. Partners Research Computing instructions screen for registering for the file transfer service that will be used by the CPET core laboratory.

Participating labs can use the following website to login to the service in order to transfer files to MGH:https://transfer.research.partners.org/courier/1000@/mail_user_login.html. In addition to basic send and receive functions, the File Manager menu tab provides a File Cabinet, Inbox and Send History for keeping track of the files you have sent and received. The core laboratory will maintain this record of file transmissions and we will encourage participating labs to do the same. Further general information about how to use this file transfer service is available at http://www.partners.org/rescomputing/content/secureFiletransfer.asp.

4.3 Core Lab Study Processing

An Excel spreadsheet will be created to track studies. Upon arrival of each study at the core laboratory the study will be logged into our database. As each study moves through the sequence of data processing, analysis, and report generation the excel sheet will be updated accordingly. An email reminder will be sent to sites if studies do not arrive in a timely fashion.

Gas exchange data will be configured uniformly in an Excel Database. Programs will then be applied to select the highest 5-breath average VO_2 during the final minute of incremental exercise. Primary breath-by-breath data will also be used to calculate anaerobic threshold, by the V slope method.[16]

5. QUALITY ASSURANCE AND QUALITY CONTROL PRACTICES

Prior to initiation of the trials, sites will be required conduct two initial tests on a "standard subject". These tests will serve to an indicator of appropriate calibration procedure, protocol adherence, and appropriate data compilation and transmission to the core laboratory. Individual CPETs will be expected to demonstrate a change in respiratory exchange ratio of >0.15 and demonstrate appropriate increases in ventilation, VO_2, and carbon dioxide production during exercise.

If, at any time, the core lab questions the quality of data that they receive from a testing site, we may require the site to perform another qualifying test. In addition to recalibration of the cycle and the metabolic cart, the core lab may request repeated studies on the "standard subject". For a normal subject, VO_2 should increase at a rate of approximately 10 ml/min/watt on a cycle ergometer. For sites that do not meet these standards, step-by-step review of each part of the exercise test, including treadmill belt speed and angle, calibration, review of potential air leaks and verification that the metabolic cart was working properly will be performed.

6. PATIENT EDUCATION FORM FOR EXERCISE TESTING

This form is as a guide only and is not mandated for use at your site

Appointment Time:_____ Date:_____

Dear _____,

You have been scheduled to have an exercise test to evaluate your exercise tolerance. You will be exercising on a special cycle or treadmill. This will begin with a 5 minute period of rest followed by 3 minutes of warm-up exercise at a very light level of work. After the warm-up the intensity of exercise will gradually increase until you are no longer able to continue. We would like you to exercise for as long as possible to make sure that we can quantify your maximum exercise performance. We anticipate that you will be exercising for about 10 minutes after the warm-up period.

If biking, we will ask you to maintain a pedal rate of 60 revolutions per minute and to not rise out of your seat during exercise testing. If walking, you must match the speed of the treadmill. You will have a mouthpiece in place that will enable us to measure the amount of oxygen that your body uses during exercise. You will be able to signal to us during testing to indicate when you will only be able to continue exercising for 1 more minute. At certain times during the test we will present you with a perceived exertion scale (see Table 6 below). You will be asked to point to the appropriate number on the scale at 2-minute intervals during the test. We will also measure your blood pressure periodically during exercise.

Pre-test instructions:
Ensure that you are well rested and adequately hydrated on the morning of the test.
Wear loose fitting, comfortable clothing that will permit you to move your legs freely.
Wear athletic shoes appropriate for exercise.
Do not engage in strenuous exercise on the day prior to the test.
Do not exercise at all within 12 hours of the test.
Do not eat anything for 3 hours before the test. Subjects with diabetes should have a light snack, as needed, to maintain adequate blood sugar levels during the test.
Avoid alcohol, caffeine, tobacco, or other stimulants within 8 hours of the test.
Take your regularly scheduled medications <u>at the same times</u> before each of your scheduled exercise tests. For example, if you take a medicine that can influence your heart rate (i.e. metoprolol, carvedilol, atenolol, verapamil, diltiazem, digoxin, albuterol) once daily at night, then take the medicine at night before each of your scheduled exercise tests.

Please answer the following questions prior to undergoing exercise testing.

*Yes	No	
☐	☐	Have you had chest pain brought on by physical activity?
☐	☐	Have you experienced dizziness, fainting, or blackouts?
☐	☐	Do you have asthma that is currently difficult to control?
☐	☐	Do you have a bone or joint problem that currently limits your ability to exercise?
☐	☐	Have you experienced fever or chills or known infection in the last 48 hours?

*For questions in which you answered yes, please indicate the time frame in which you experienced these symptoms:_____

Table 6. Borg 10-grade new rating scale with ratio properties to be applied to the degree of shortness of breath experienced during exercise.

SCALE	Descriptive Term	Level of Exertion
0	Nothing at all	
0.5	Very, very weak	Just noticeable
1	Very weak	
2	Weak	Light
3	Moderate	
4	Somewhat strong	
5	Strong	Heavy
6		
7	Very Strong	
8		
9		
10	Very, very strong	Almost Max

7.PROTOCOL SPECIFIC WORKSHEETS

7.1 Qualification Testing, Normal Subject, CYCLE ERGOMETRY

Referring Center Name and Number:_____
PI Name: _____
Date, Time, Study description (A or B):_____
Gas Exchange Equipment and Software Manufacturer:_____

Table 7. CPET exercise worksheet for standard subject performing cycle ergometry

Elapsed Time (min)	Work Rate Increase at 20 W/min	RPM (min^{-1})	HR	O_2 sat (%)	SBP (mmHg)	DBP (mmHg)	Borg Dyspnea (1-10)
-1 (Pre)	Rest	0					
0-5	Rest	0					
5-8	0 (unloaded)	60					
8-9	20	60					
9-10	40	60					
10-11	60	60					
11-12	80	60					
12-13	100	60					
13-14	120	60					
14-15	140	60					
15-16	160	60					
16-17	180	60					
17-18	200	60					
18-19	220	60					
19-20	240	60					
20-21	260	60					
21-22	280	60					
>22	+20/min	60					
Peak: t_____	**W_____**						
Rec. 1 min		0					
Rec. 2 min		0					
Rec. 3 min		0					
Rec. 4 min		0					
Rec. 5 min		0					
Rec. 6 min		0					
Rec. 7 min		0					
Rec. 8 min		0					
Rec. 9 min		0					
Rec. 10 min		0					

Rec. indicates recovery, W indicates watts, HR indicates heart rate, SBP indicates systolic blood pressure, DBP indicates diastolic blood pressure.

7.2 Qualification Testing, Normal Subject TREADMILL testing

Sites will only complete this protocol if a cycle ergometer is not available)

Cardiopulmonary Exercise Worksheet

Referring Center Name and Number:_____

PI Name: _____

Date, Time, Study description (A or B):_____

Gas Exchange Equipment and Software Manufacturer:_____

Table 8. CPET exercise worksheet for standard subject performing treadmill ergometry

Elapsed Time (min)	Work Rate (Increase 20W/min)	Grade (%)	Speed (mph)	HR (bpm)	O_2 sat (%)	SBP (mmHg)	DBP (mmHg)	Borg Dyspnea (0-10)
-1 (Pre)	Rest	0	0					
0-5	Rest	0	0					
5-8	10	4	0.8					
8-9	30	9.5	1.1					
9-10	50	12.5	1.3					
10-11	70	14.5	1.6					
11-12	90	16.5	1.8					
12-13	110	17.5	2.1					
13-14	130	18.5	2.3					
14-15	150	19	2.6					
15-16	170	20	2.8					
16-17	190	20.5	3.1					
17-18	210	21	3.3					
18-19	230	21.5	3.5					
19-20	250	22	3.7					
20-21	270	22.5	3.9					
21-22	290	23	4.1					
>22		↑0.5/min	↑0.2/min					
Peak: t____								
Rec. 1 min	10	0	1.1					
Rec. 2 min	Rest	0	0					
Rec. 3 min	Rest	0	0					
Rec. 4 min	Rest	0	0					
Rec. 5 min	Rest	0	0					
Rec. 6 min	Rest	0	0					
Rec. 7 min	Rest	0	0					
Rec. 8 min	Rest	0	0					
Rec. 9 min	Rest	0	0					
Rec. 10 min	Rest	0	0					

Rec. indicates recovery, W indicates watts, SBP indicates systolic blood pressure, DBP indicates diastolic blood pressure, t indicates time elapsed at cessation of exertion.

7.3 IRONOUTHF 10 Watt Ramp CYCLE ERGOMETER PROTOCOL

Table 9. CPET exercise worksheet for subjects performing 10W/min ramp cycle ergometry

HCM♥NET CPET CORE PROTOCOL SYNOPSIS AND SUPPLMENTAL DATA SHEET

Referring Center Name and Number:		**Hemoglobin**	
Subject/Test NAME:		**Time of study med**	
Date and time at start of exercise:		**Seat height (cm)**	
Technician of record:		**FEV1**	
Reason for cessation of exercise		**FVC**	

Elapsed Time(min)	Work Rate Increase 10 W/min	Target Pedal Rate (rpm)	Actual Pedal Rate (rpm)	HR (bpm)	O_2 sat (%)	SBP (mmHg)	DBP (mmHg)	Borg Dyspnea (1-10)
-1 (Pre)	Rest	0						
0-5	Rest	0						
5-8	0	60						
8-9	10	60						
9-10	20	60						
10-11	30	60						
11-12	40	60						
12-13	50	60						
13-14	60	60						
14-15	70	60						
15-16	80	60						
16-17	90	60						
17-18	100	60						
18-19	110	60						
19-20	120	60						
20-21	130	60						
21-22	140	60						
22-23	150	60						
23-24	160	60						
24-25	170	60						
>25	10/min	60						
Time:	**W:**							
Rec. 1 min	5	30						
Rec. 2 min	rest	0						
Rec. 3 min	rest	0						
Rec. 4 min	rest	0						
Rec. 5 min	rest	0						
Rec. 6 min	rest	0						
Rec. 7 min	rest	0						
Rec. 8 min	rest	0						
Rec. 9 min	rest	0						
Rec. 10 min	rest	0						

*Recordings should be made during the last 15 sec of each minute

7.4 IRONOUT: TREADMILL PROTOCOL. Designed to mirror cycle ergometry protocol with a ≈10W/min incremental protocol

Table 10. CPET exercise worksheet for subjects performing 10W/min treadmill ergometry assuming subject weight <= 80 kg

HCM♥NET CPET CORE TREADMILL PROTOCOL SYNOPSIS AND SUPPLMENTAL DATA SHEET					
Referring Center Name and Number:			**Hemoglobin**		
Subject/Test NAME:			**Time of study med**		
Date and time at start of exercise:					
Technician of record:			**FEV1**		
Reason for cessation of exercise			**FVC**		

Elapsed Time (min)	Work Rate (W)	Speed (mph)	Grade (%)	HR (bpm)	O_2 sat (%)	SBP (mmHg)	DBP (mmHg)	Borg Dyspnea (1-10)
-1 (Pre)								
0-5	Rest	Rest	0					
5-8	5	0.8	2					
8-9	15	1	5					
9-10	24	1.1	7					
10-11	33	1.3	8.5					
11-12	43	1.5	9.5					
12-13	51	1.6	10.5					
13-14	60	1.8	11					
14-15	70	2	11.5					
15-16	80	2.2	12					
16-17	88	2.3	12.5					
17-18	100	2.5	13					
18-19	111	2.7	13.5					
19-20	123	2.9	14					
20-21	137	3.1	14.5					
21-22	150	3.3	15					
22-23	165	3.5	15.5					
23-24	180	3.7	16					
24-25	195	3.9	16.5					
>25		+0.2/min	+0.5/min					
Peak: t_____								
Rec. 1 min	5	1.0	0					
Rec.2 min	0	rest	0					
Rec. 3 min	0	rest	0					
Rec. 4 min	0	rest	0					
Rec. 5 min	0	rest	0					

Rec. indicates recovery, W indicates watts, HR indicates heart rate, SBP indicates systolic blood pressure, DBP indicates diastolic blood pressure, t indicates time of cessation of exercise.

Table 10b. CPET exercise worksheet for subjects performing treadmill ergometry
Designed to mirror cycle ergometry protocol with a ≈10W/min incremental protocol assuming subject weight > 80 kg

HCM♥NET CPET CORE TREADMILL PROTOCOL SYNOPSIS AND SUPPLMENTAL DATA SHEET					
Referring Center Name and Number:			**Hemoglobin**		
Subject/Test NAME:			**Time of study med**		
Date and time at start of exercise:					
Technician of record:			**FEV1**		
Reason for cessation of exercise			**FVC**		

Elapsed Time (min)	Work Rate (W)	Speed (mph)	Grade (%)	HR (bpm)	O_2 sat (%)	SBP (mmHg)	DBP (mmHg)	Borg Dyspnea (1-10)
-1 (Pre)	0	Rest	0					
0-5	0	Rest	0					
5-8	5	0.8	1.5					
8-9	16	1	4					
9-10	24	1.1	5.5					
10-11	36	1.3	7					
11-12	44	1.5	7.5					
12-13	50	1.6	8					
13-14	60	1.8	8.5					
14-15	70	2	9					
15-16	82	2.2	9.5					
16-17	90	2.3	10					
17-18	98	2.5	10					
18-19	111	2.7	10.5					
19-20	125	2.9	11					
20-21	140	3.1	11.5					
21-22	155	3.3	12					
22-23	164	3.5	12					
23-24	181	3.7	12.5					
24-25	191	3.9	12.5					
>25		+0.2/min	+0.5/min					
Peak: t____								
Rec. 1 min	5	1	0					
Rec.2 min	0	rest	0					
Rec. 3 min	0	rest	0					
Rec. 4 min	0	rest	0					
Rec. 5 min	0	rest	0					

Rec. indicates recovery, W indicates watts, SBP indicates systolic blood pressure, DBP indicates diastolic blood pressure, t indicates time elapsed upon cessation of exercise.

8. IRONOUT-HF CPET CORE LAB PERSONNEL

Primary Contact:

Core Laboratory Manager	Diane Cocca-Spofford RN Tel: 617-726-8228 E-mail: dcoccaspofford@partners.org
Mailing Address:	MGH Core CPET Laboratory GRB 800,Cardiology Division Massachusetts General Hospital 55 Fruit St, Boston, MA 02114

Additional Laboratory Personnel Contact Information

Core Laboratory Director	Gregory D. Lewis MD Tel: 617-724-6158 Email: glewis@partners.org
Core Laboratory Staff	Marc J. Semigran Tel: 617-726-8662 Email: msemigran@partners.org
Core Laboratory Chief Technician	Paul Pappagianopoulos MEd Tel: 617-724-7825 Email: ppappagianopoulos@partners.org
Core Laboratory Director of Information Techonology	Eugene Pomerantsev MD, PhD Tel: 617-724-5585 Email: epomerantsev@partners.org
Core Laboratory Technical Support Staff	Aaron Eisman BA Tel: 617-643-9133 E-mail: aeisman@partners.org

APPENDIX 1: Form 001 IRONOUT-HF CPET Core Lab

Site Qualification Procedures Checklist:

☐ Verify your site's compliance with equipment requirements and calibration procedures.

☐ Email Diane Cocca-Spofford RN at dcoccaspofford@partners.org and CC glewis@partners.organd indicate the following:

a. Responsible CPET lab staff member who will primarily interact with the Core Laboratory with email address and contact information.
b. Exercise modality that will be used for the clinical trials (cycle or treadmill)
c. Metabolic cart manufacturer and software program that will be used.

☐ Accept emailed invitation from Diane Cocca-Spofford to join the Partners Research Computing Secure Files Transfer Service (see Section 4 for detailed instructions).

☐ Submit two incremental symptom-limited CPET qualifying tests on a "standard normal subject" (see Section 7, protocol specific worksheets) via the Partners Computing Secure Files Transfer Service.

CPET Procedures Checklist:

☐ Print and distribute patient education forms (Section 6) to subjects at least 48 hours prior to testing and assess compliance with instructions upon the subject's arrival to the laboratory.

☐ Obtain the subject's study ID number from your site's research coordinator.

☐ Assess CPET contraindications (Table 1).

☐ Print Borg Score Table for use during CPET (section 6).

☐ Perform spirometry and record results in metabolic cart software program.(optional)

☐ Configure gas exchange data output according to Table 5.

☐ Enter current hemoglobin level and cycle seat height into CPET electronic file.

☐ Complete CPET according to the appropriate protocol (sections 3 and 7), integrate all study data into a single CPET file, and name the file according to section 4.1.

☐ Save a backup copy of the CPET file on a disk that will be maintained in individual CPET laboratories and transmit the electronic file to the core laboratory.

☐ Transmit data file to the core laboratory.

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