ECHOCARDIOGRAPHIC STUDY OF LATINOS (ECHO-SOL)

Echocardiography Reading Center Manual of Operations

Carlos J. Rodriguez MD,MPH, FACC
ECHO-SOL Director
Echocardiography Reading Center PI

Table of Contents

Wake Forest School of Medicine

September 23, 2015

Dear Participating Sites,

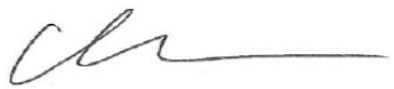
We are pleased to provide you with the Manual of Procedures for ECHO-SOL. In this protocol, you will find all of the information you will need to gather the necessary cardiovascular ultrasound data. Your site coordinators and cardiovascular ultrasound technologists should review the protocol upon receipt to familiarize themselves with the requirements. Any questions should be addressed as soon as possible to facilitate a clear understanding of protocol prior to the arrival of your first participant.

As a participating site, your main responsibility is to perform complete and accurate cardiovascular ultrasound exams while strictly adhering to the procedure outlined in this protocol. It is critical that the integrity of the study is maintained through each study site by adherence to this Manual of Procedures and conformity between all the sites. Every site will be asked to electronically ship digitally archived images along with data tracking sheets to the Cardiac Echocardiography Reading Center at Wake Forest School of Medicine and retain copies of all data locally. Each clinic is required to obtain and deliver a "test case" at first opportunity; please see further instructions within this protocol manual.

Please note that the protocol for ECHO-SOL may differ from the scanning protocol used in your clinical practice. Your site **must** follow this standardized protocol in order to maintain the integrity of the study results. The protocol for this study is designed for research purposes only and does not substitute for a complete echocardiographic examination as might be needed for the clinical evaluation or management of the patient.

Feel free to contact me or the ECHO-SOL staff if we can lend assistance in any way. We are always available to answer any questions and to guide you through the entire process. Thank you, in advance, for your commitment to the study and we look forward to working with you.

Sincerely,



Carlos Jose Rodriguez MD, MPH, FACC
Associate Professor of Epidemiology and Medicine
Wake Forest University School of Medicine
Medical Center Boulevard
Winston-Salem, NC 27157

I. Overall Study Aims and Processes

The overall goal of ECHO-SOL (Echocardiographic Study of Latinos/Hispanics), is to establish a unique cohort that will permit study of cardiac structure and function in Hispanic adults. ECHO-SOL will be an ancillary study to the NIH-funded Hispanic Community Health Study – Study of Latinos (HCHS-SOL), a population-based study of 16,000 Hispanics. ECHO-SOL will consist of 1,800 participants already enrolled in HCHS-SOL across four US sites (Bronx; Chicago; San Diego; Miami), utilizing a balanced enrollment design to obtain near-equal numbers of Hispanic subgroup representation. Participants will be characterized with echocardiograms using state-of-the-art techniques (tissue Doppler imaging and strain imaging analysis). The primary outcomes are the presence of abnormal cardiac structure (as defined by LVH), abnormal systolic function (as defined by LV ejection fraction) and abnormal diastolic function (as defined by a validated algorithm). Secondary outcomes of interest will include LV geometry, as well as LV systolic and diastolic strain.

ECHO-SOL will provide for the first time, the largest dataset of echocardiographic parameters focused solely on US Hispanics with strong subgroup representation. This study will allow for the determination of LVH prevalence among Hispanics and be the first to untangle the relative importance of both the traditional and psychosocial / socioeconomic determinants of cardiac structure and function.

The Imaging Core Laboratory for this study will be located at Wake Forest University School of Medicine and directed by Dr. Carlos J. Rodriguez. For the proposed study, the functions of the research echocardiography core laboratory will include the following:

1. Work collaboratively with the Field Centers and the CCSC to implement the protocol for the data acquisition, quality control and assurance, and information abstraction of the echocardiograms.
2. Train and certify echocardiography technicians for each Field Center according to protocol.
3. Conduct a pretest of the echocardiography protocol, including quality assurance activities.
4. Perform readings and information abstractions of echocardiograms in a timely manner during the pilot phase and throughout the study period.
5. Track the receipt, reading, and storage of echocardiograms.
6. Implement long-term archiving of echocardiograms and timely creation of backup copies.
7. Maintain confidentiality and security of the data files.
8. Transmit results of the echocardiography reading and quality control assessments at all phases of the study to the CCSC on a regular and timely basis.

OBJECTIVES	**Echocardiography**	• The objective of the ECHO-SOL is to describe and quantify perturbations in cardiac size and function across the spectrum of Hispanics in the HCHS-SOL cohort. • Echocardiographic examinations will be performed to estimate myocardial structure and performance including but not limited to: left and right ventricular systolic function, left ventricular end diastolic volume (LVEDV), left ventricular end systolic volume (LVESV), left ventricular mass, left atrial size, LV diastolic function, mitral inflow pulsed wave Doppler (E wave, A wave), isovolumic relaxation time (IVRT).
	Cardiac Imaging Core Lab	• To provide high quality reproducible quantitative analysis of study echocardiograms • Interact with field centers for issues related to study deficiencies, chronic poor quality studies and other issues related to overall site performance.
	Site Instruction Manual	• To instruct field centers on how to perform and send echocardiograms to the Echo Core Lab.

ROLES AND RESPONSIBILITIES	**Field Centers**	• Perform high-quality study echocardiograms per the protocol contained in this document
	HCHS-SOL Coordinating Center	• Ensure that the Echo Core Lab stays informed of study-wide changes and updates as the study progresses. • Provide oversight and support, as required, for the entire process
	Cardiac Imaging Core Lab	• Receive, review and analyze study echocardiograms.. • Train and certify each field center sonographer. • Provide field centers feedback on poor quality echocardiograms, and queries for technical / process improvement. • Serve as a resource for sites regarding echo-related questions.

III. Echocardiogram Protocol: Required Views

OBJECTIVES

- The primary objective of the initial echo-Doppler examination is to establish baseline cardiac anatomy and function with emphasis on left ventricular size and function.
- Two-dimensional ultrasound imaging system with pulsed, continuous wave and color Doppler capability, and permanent recording capabilities are required. The only accepted storage format is Digital Imaging and Communications in Medicine (DICOM) standard digital.
- Each site should have transducers with both fundamental and harmonic capability with frequency ranges that are suitable for most adult subjects (approximately 2 to 7 MHz).

EXAMINATION PREPARATION, POSITIONING, AND GENERAL PROCEDURES

- Consent will have been obtained by SOL study coordinators before the participants come to the imaging site. Prior to starting the study, the sonographer will provide the patient with an overview of the echocardiographic study (duration, general procedure, etc).
- Patients will typically be studied while they are in the left lateral decubitus position although occasionally the best images will be obtained with the patient supine. Each sonographer is responsible for ensuring optimal patient position.
- EKG leads will be applied and the EKG control setting of the machine optimized to ensure a high quality EKG with adequate amplitude QRS complex for reliable digital capture (lead II equivalent preferred).
- All echo images will be sent de-identified (without any *individually identifiable* patient health information) to the CERC at WFSM. All exams must be appropriately labeled and accompanied by a completed tracking form. Enter ECHO SOL in the [**first name**] field, the study site in the [**last name**] field, the HCHS/SOL ID in the [**MRN**] field, and exam date as specified on the echo tracking form. Please measure and note the patient's blood pressure, height, weight, gender, and age on the patient ID screen or as an annotation on one or more images.

- All views will be acquired with captures as follows:

  Sinus rhythm (up to 90 bpm): 3 beat capture
  Sinus rhythm (≥ 90 bpm): 5 beat capture
  Frequent atrial or ventricular ectopy: 3 second capture
  Atrial fibrillation or flutter: 5 second capture

- It is essential that the view be optimized and stabilized before recording. Breath holding during recording is not required unless necessary to ensure a stable high quality image.
- The spatial resolution of the images should be optimized using the highest frequencies capable of providing adequate penetration.
- It is important that the same machine type (make, model, pre and post processing settings) and transducer be used on all initial studies.
- For the parasternal long axis view used for measurements, fundamental imaging should be attempted. If images are inadequate, harmonic imaging may be used. Harmonic imaging may be used for all other views.
- The sweep speed for all spectral Doppler and M-mode recordings shall be 100mm/sec.
- In all spectral Doppler interrogation gain and filters should be adjusted to reduce excess noise

EXAMINATION SEQUENCE

Parasternal Long Axis

- Take a 2D loop of PSLAX with increased (deep) depth for effusion
- Heart must be horizontal. One clip obtained at decreased depth optimizing the left ventricle (LV) (show pericardium).
- Zoom LVOT / AV (Adjust the box size big enough to include some space before and after the valve)
- Color Doppler AV
- Color Doppler MV (Adjust the color box size to include some space before and after the MV; color box should cover LA)

Parasternal Short Axis (please ensure on-axis views)

- Basal Parasternal Short Axis (PSAX) (at the aortic valve level):
  - 2D loop PSAX AV level showing TV, AV (show all AV leaftlets) and PV

- Evaluation of TV performed (can go off axis to get better TV inflow) color Doppler tricuspid valve (TV)
- One clip CW TV for TR (try to adjust TR parallel to beam)
- Evaluation of PV; Acquire a 2D loop of PV (can be off axis to get well defined PV). Optimize the PV annulus for measurement of the pulmonary annular diameter
- Color Doppler PV
- PW Doppler sample at the level of the pulmonary valve (PV) leaflet tips to measure pulmonary velocity time interval (VTI).
- One clip of CW PV. Opening and closing transients of the pulmonary valve should be recorded.

PSAX continued for left ventricle:

- One clip at mitral valve (MV) level (base)
- One clip mid-ventricle (mid-papillary muscle level)
- One clip m-mode recording of the LV just below the tips of the mitral leaflets.
- One clip at the level of LV apex

4 Chamber Apical

- One clip demonstrating all 4 chambers with good endocardial definition for left atrium (LA) & left ventricle (LV) volume measurements
- One clip at Decreased depth and focus on LV
- Color MV
- PW Doppler of mitral inflow at mitral leaflet tips
- CW of mitral inflow
- PW Doppler of right upper pulmonary vein flow. The sample volume should be placed at least 1 cm within the pulmonary vein, if possible
- Tissue Doppler Interval (TDI) of septal mitral annulus
- Tissue Doppler Interval (TDI) of lateral mitral annulus
- One clip optimizing RV full screen (either change depth or zoom)

- One clip color Doppler TV for tricuspid regurgitation
- CW TV for TR
- TDI for anterior tricuspid annulus
- M-mode of anterior tricuspid annulus.
- 2D loop RV full screen (either change depth or zoom to optimize RV)

Anteriorly angulated 4-chamber view: Apical 5 chamber

- 2D loop of AP5 chamber
- Color AV
- PW LVOT (sample volume positioned such that closing artifact but not opening artifact of the valve is visible)
- CW of the AV
- PW in between mitral and aortic inflow for IVRT (Isovolumic Relaxation Time) measurement

2-Chamber Apical

- One clip LV & LA full screen (either change imaging depth or zoom) with good endocardial definition for LA & LV volume measurements
- Zoom LV or decrease the depth so LV is in focus
- Zoom LA

3-Chamber Apical

- One clip LV & LA & AV (optimize either depth or zoom)
- One clip Color MV
- One clip Color AV (CW only if not acquired in 5-chamber view)

SUMMARY OF ECHO PROTOCOL

A. Blood pressure	
☑ *Brachial blood pressure*	♦ Measure BP just prior to the echo examination
B. Parasternal Position	
☑ *Parasternal long axis*	♦ 2D imaging (at deep depth) ♦ 2D imaging (at shallow depth till pericardium) ♦ Zoom AV ♦ Color Doppler AV and MV in separate views
☑ *Parasternal short axis – Aortic valve level*	♦ 2D imaging ♦ Color Doppler TV ♦ CW Doppler TV ♦ 2D imaging focus on PV (can be off axis for well-defined PV) ♦ Color Doppler PV ♦ PW and CW Doppler PV
☑ *Parasternal short axis – Mitral valve level*	♦ 2D imaging
☑ *Parasternal short axis – Mid-Papillary muscle level*	♦ 2D imaging ♦ M-mode
☑ *Parasternal short axis – LV apex*	♦ 2D imaging
C. Apical Position	
☑ *Apical 4 chamber view– focused on the LV*	♦ 2D imaging AP4 ♦ Zoom or Decreased depth and focus on LV ♦ Zoom LA ♦ Color Doppler MV ♦ PW and CW Doppler MV ♦ PW Doppler Pulmonary vein ♦ TDI of septal and lateral mitral annulus
☑ *Apical 4 chamber – focused on the RV*	♦ 2D imaging RV(optimize RV, can go off axis to optimize view) ♦ Color Doppler of TV/TA ♦ CW Doppler of tricuspid regurgitation ♦ TDI of anterior tricuspid annulus ♦ M-mode anterior tricuspid annulus
☑ *Apical 5 chamber view*	♦ 2D imaging ♦ Color Doppler AV ♦ PW and CW Doppler of AV ♦ PW of LVOT/aortic outflow and mitral inflow (for IVRT)
☑ *Apical 2 chamber view*	♦ 2D imaging ♦ Zoom LV or decrease depth and focus on LV ♦ Zoom LA
☑ *Apical 3 chamber view*	♦ 2D imaging ♦ Color Doppler AV and MV

Detailed Review of Protocol Required Views

A. Brachial Blood Pressure

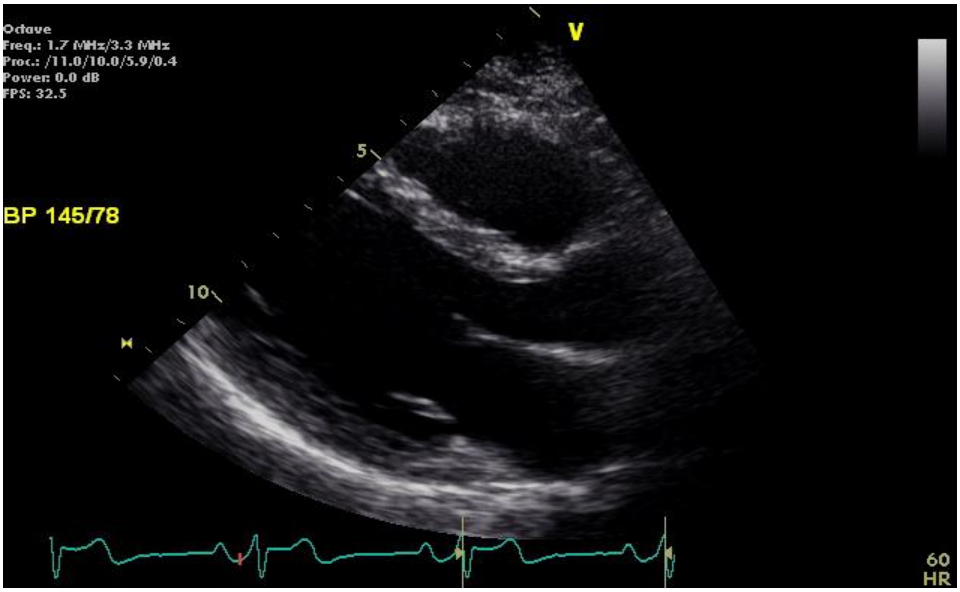
Just prior to the start of the echocardiographic examination, brachial blood pressure is measured. The subject's blood pressure should be taken **just prior** to the echocardiogram and after the subject has been resting for 5 minutes. Blood pressure should be performed at baseline in both arms. The highest reading should be recorded and subsequent measure should be done on the arm with the highest reading. Be sure to record the blood pressure and initial heart rate on the Echo Tracking Form.

B. Parasternal Views

Two parasternal views will be obtained:

- Parasternal long axis view
- Parasternal short axis view at 4 levels as detailed below (section B.2.)

B.1. Parasternal Long Axis View



Parasternal Long Axis

In the ideal echocardiographic "window" for the long axis:

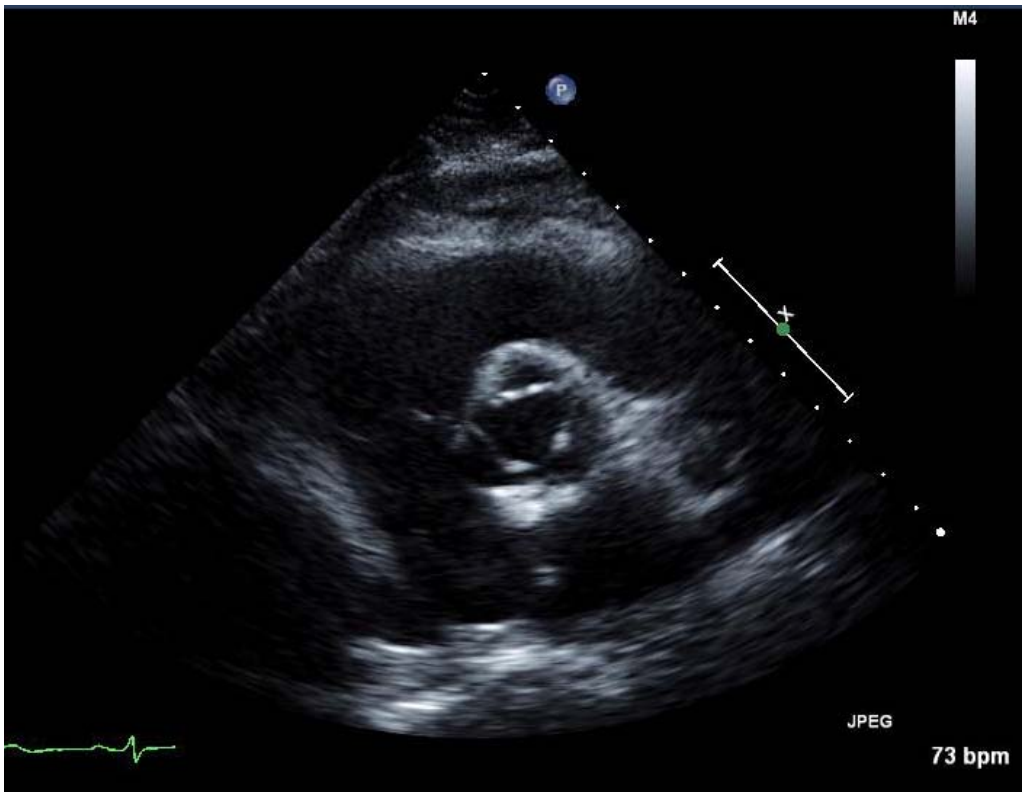
- The proximal interventricular septum is horizontal and continuous with the aortic root.
- The anterior and the posterior mitral valve leaflets, and the right and non-coronary aortic valve leaflets are visible.
- The left ventricular apex is not visualized.
- Most importantly: the LV endocardium at the septum and the posterior wall are well delineated.

B.2. Parasternal Short Axis View

Parasternal short axis view will be obtained at four levels:

1. At the aortic valve level with the RVOT and pulmonic valve visible.
2. At mitral valve when both anterior and posterior mitral valve leaflets are visualized.
3. At the mid-papillary muscle level with the papillary muscles visible.
4. At the left ventricular apex.

B.2.i. Aortic Valve Level



Acquire both 2D imaging and color Doppler of this view.

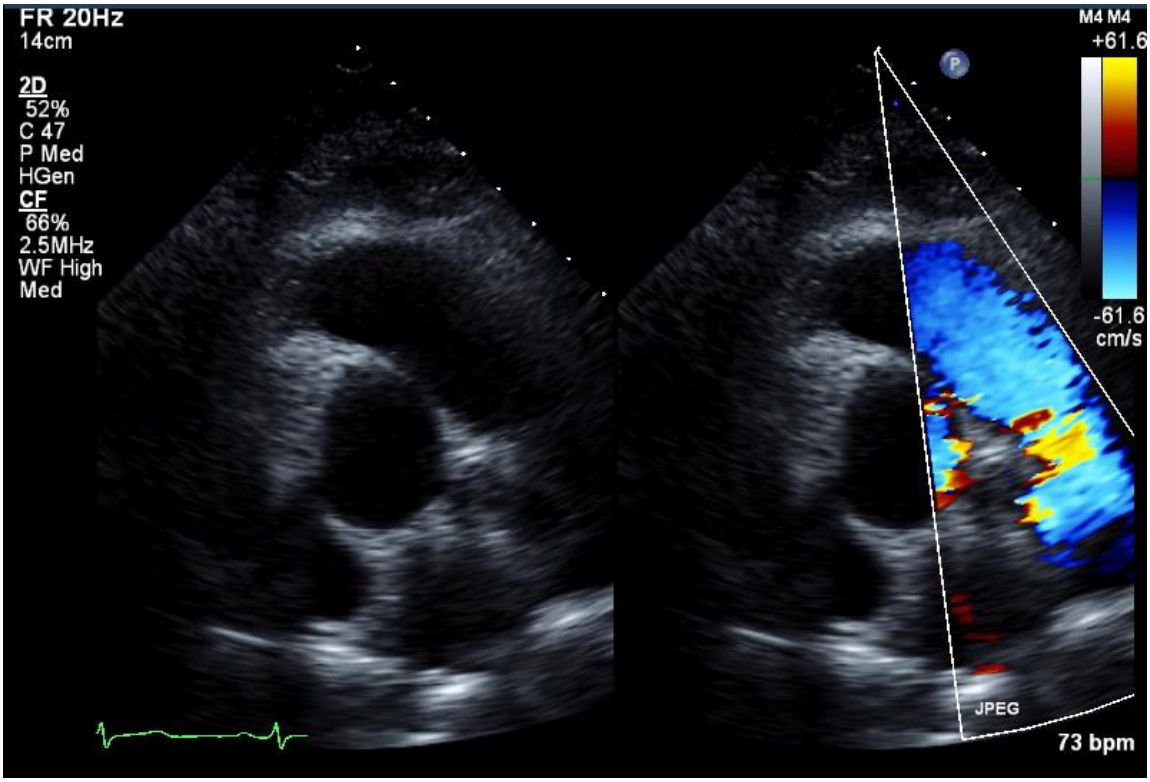
- 1 clip with all 3 cusps of the aortic valve visible, with a clear upside down triangle pattern during systole.
- The tricuspid valve and interatrial septum are visible.

Evaluation of TV performed;

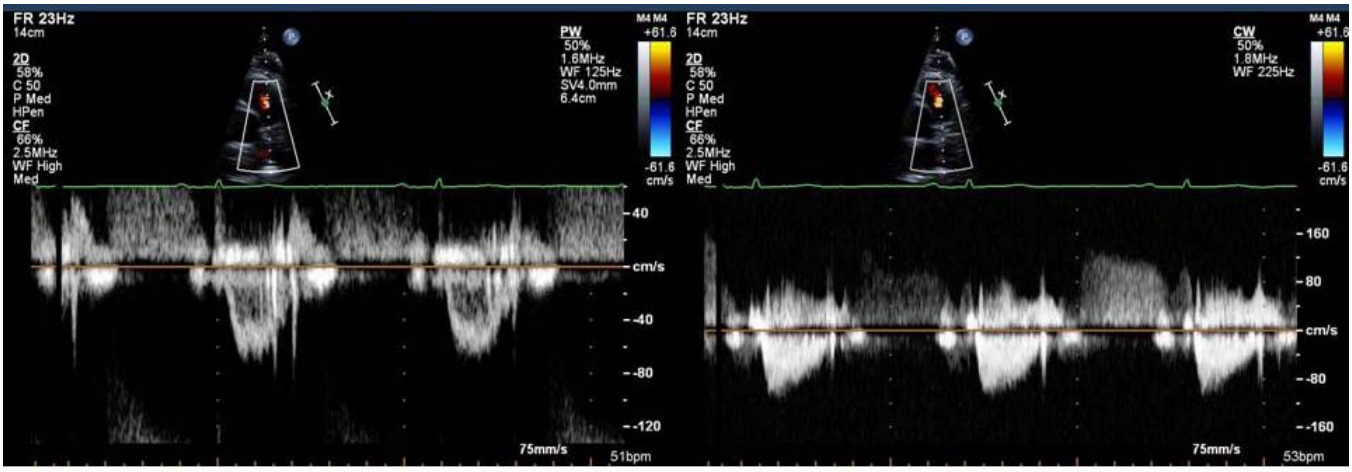
- 1 clip with and without color flow Doppler. Pulsed 1 clip color Doppler tricuspid regurgitation
- 1 clip CW TR jet

From the parasternal short axis view at the aortic valve level, the following additional images will also be obtained:

- **PSAX view focused on the right ventricular outflow tract and pulmonic valve**



- **Continuous and pulsed wave spectral Doppler of RVOT and trans-pulmonic flow**
- **Optimize for systolic and diastolic flow**

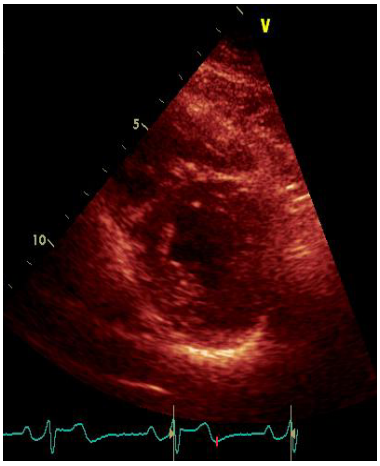


PW Doppler **CW Doppler**

B.2.ii. Mitral valve level, mid-papillary level, and apical level

Mitral valve level

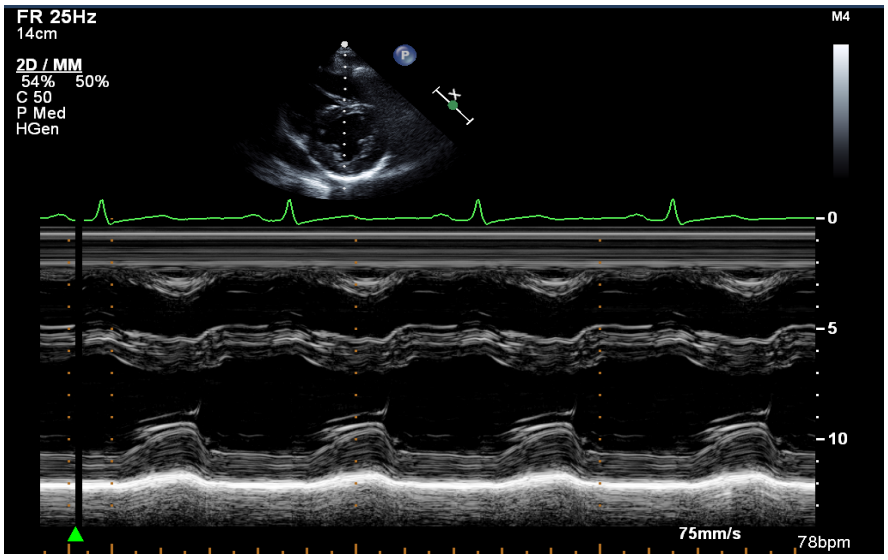
Mid-papillary level



Apical Level

In the ideal echocardiographic "window" for the short axis:

- In the absence of prior infarction, the left ventricle should have a circular shape in the short axis – an elliptical shape suggests off-axis/tangential cut through the ventricle.
- Use internal LV landmarks to ensure imaging at consistent planes in the short axis: visualization of the anterior and posterior mitral leaflets for the mitral valve leaflets; visualization of both papillary muscles for the mid-papillary level
- For the quantification of left ventricular mass, M-mode recordings of the left ventricle should be obtained with the ultrasound beam at or just below the tips of the mitral valve leaflets. Minimal valvular apparatus should be visible:



C. Apical Views

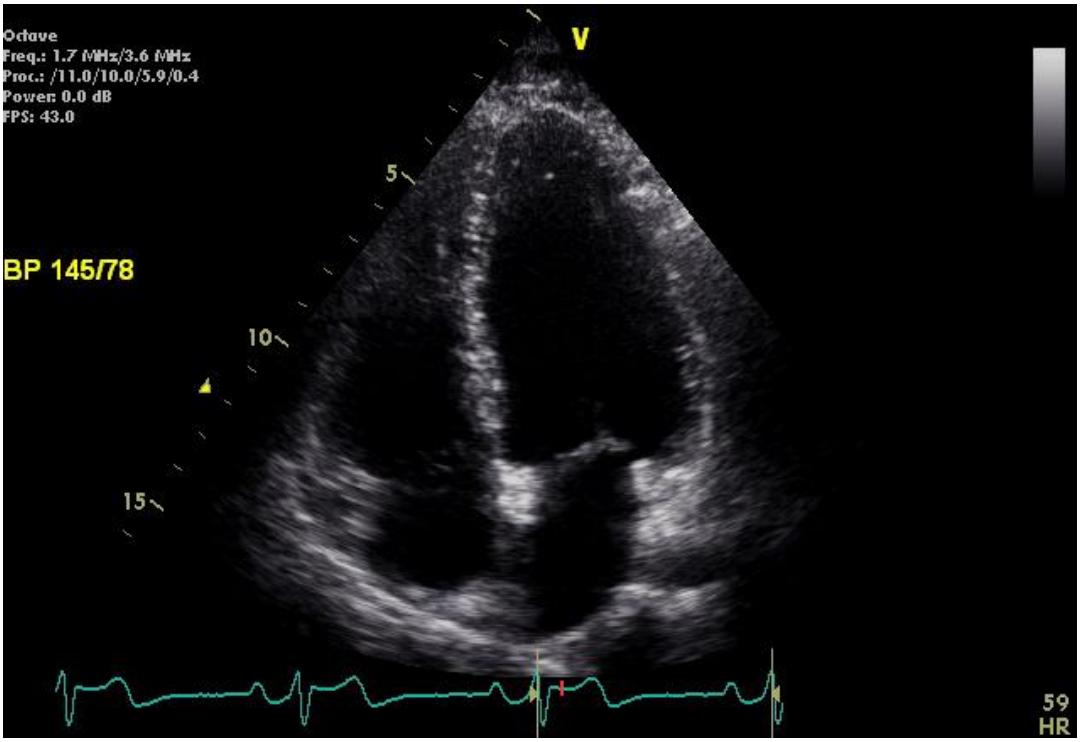
The following apical views will be obtained:

- The standard apical four-chamber focused on the LV
- The apical four-chamber dedicated to optimal imaging of the RV
- The five-chamber view
- The two-chamber view
- The three-chamber view

At the Reading center, left ventricular and atrial areas and volumes will be measured from these views (i.e. using Simpson's method). Therefore, in all apical views, special attention should be paid to properly align the image and capture the left ventricle and atrium in full. Avoid either foreshortening or elongating the chambers by transducer angulation.

The entire LV endocardium must be within the sector scan in both end-diastole and end-systole. The most difficult areas in which to visualize the endocardium are usually the apex and the lateral LV free wall and particular attention must be paid to these areas.

C.1. Apical 4-Chamber View



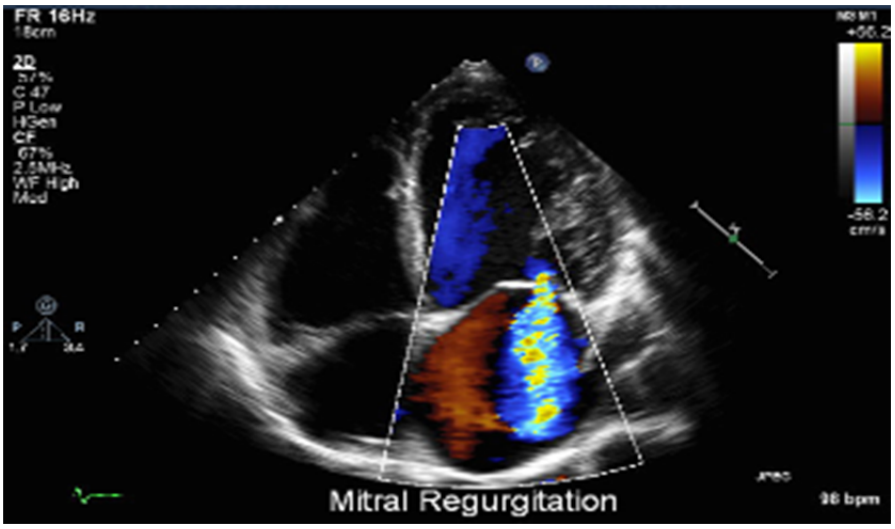
Apical 4-Chamber

- Obtain 1 clip optimizing visualization of the left ventricle during systole and diastole. Obtain a second clip optimizing visualization of the left atrium throughout systole and diastole.

  In the ideal echocardiographic 'window' for the Apical 4-Chamber view:

- Maximize LV length and be careful not truncate the true long axis.
- The entire LV endocardium must be within the imaging sector in both end-diastole and end-systole. Pay special attention to the apex and the lateral LV free wall, which are often the most difficult areas to visualize.
- Be sure to properly align the image and capture the left atrium in full. Avoid any foreshortening of the chamber.

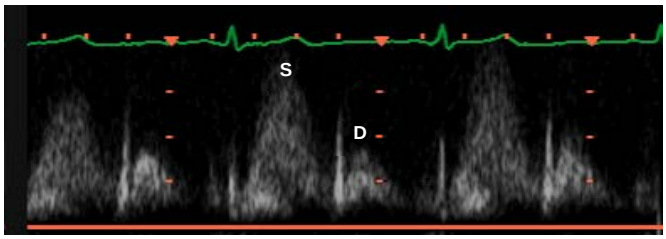
C.1.i. Color Flow Doppler of Mitral Regurgitation



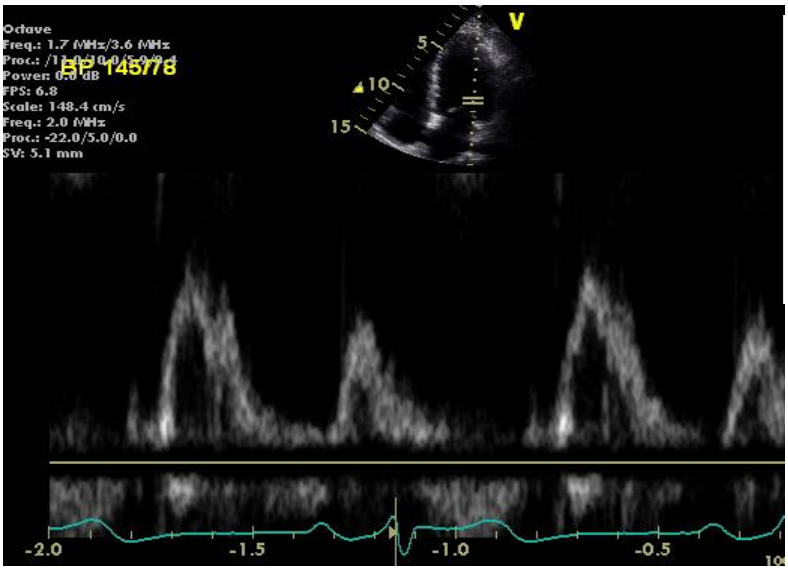
- Adjust color Doppler sample sector over the mitral valve and include the entire LA cavity. To optimize frame rate, keep the color sector scan as narrow as possible, while including the entire LA. The Nyquist limit should be set at 60 cm/s.

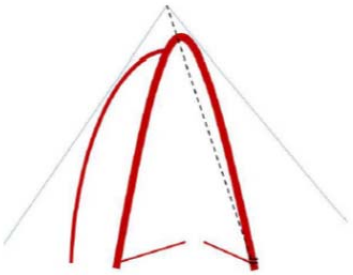
Pulsed Wave Doppler Interrogation of Pulmonary Venous Flow

Measurement of pulmonary venous flow involves placement of the pulsed wave Doppler sample volume 1 to 2 cm into the lumen of the right upper pulmonary vein and recording systolic and diastolic inflow velocities. In the image below, S stands for systolic velocity, and D stands for diastolic velocity.



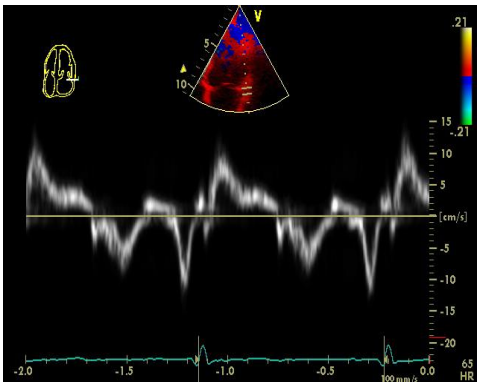
C.1.ii. Spectral Doppler of Mitral Inflow



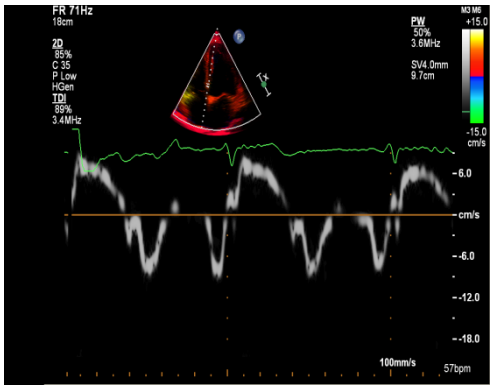


- From the apical four chamber view record the mitral inflow velocity curve with the pulsed-wave Doppler sample volume positioned at the tips of the mitral leaflets during quiet respiration. Adjust the baseline and Doppler scale to visualize the peak E and A wave velocities.

C.1.iii. Tissue Doppler Imaging (TDI) of Mitral Annulus (lateral and septal)



Lateral mitral annulus



Septal mitral annulus

- Decrease image depth (to include the LV and a small part of the LA, ideal depth approximately 16 cm) and optimize the 2D image for the LV, focusing on the lateral wall and the mitral annular region.

- Adjust the image to orient the motion of the lateral wall parallel to the cursor. Both gains and filter settings should be set low (100 Hz or less) to obtain the best images.

- Initiate 2D color DTI and **position the sample volume on the ventricular side of the lateral mitral annulus at the junction of the LV wall with the mitral annulus** of the lateral myocardial segment; the myocardium should stay within the sample volume for as much of the cardiac cycle as possible.

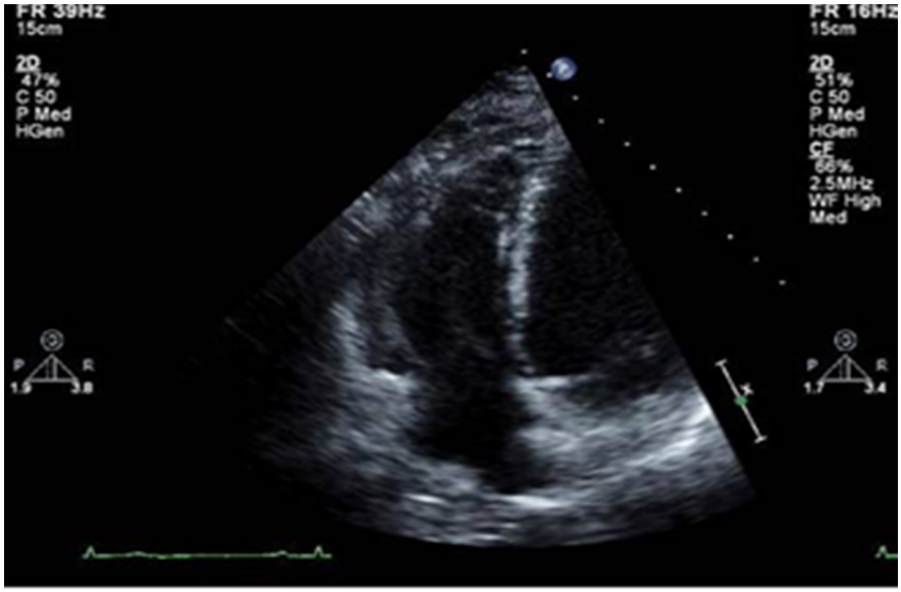
- Before the data is acquired, check that only the region to be sampled is moving through the sample volume.

- Switch to PW spectral DTI and set the scale to 20 cm/sec with a sweep speed of 100 mm/sec.

- Once a clear pattern is obtained, record at least 2 beats during quiet respiration (or preferably at end-expiration).

- Repeat this process for the septal mitral annulus

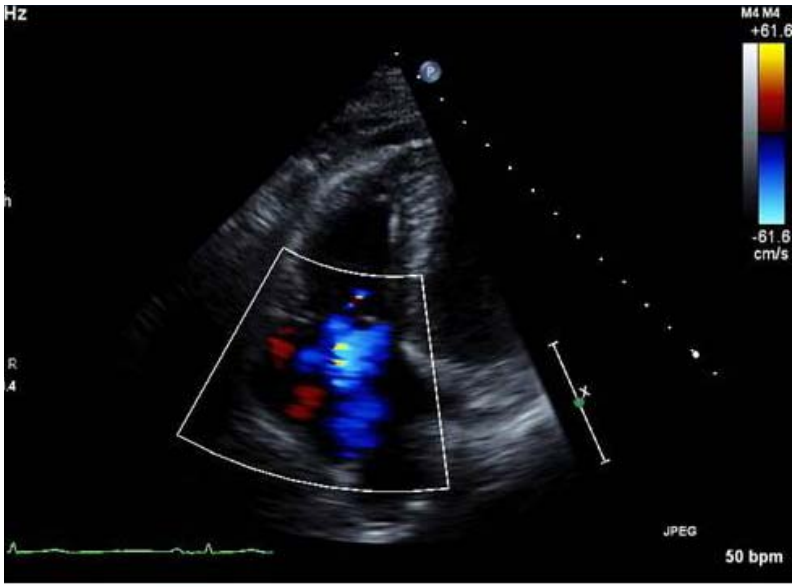
C.2. Apical 4-Chamber View (Focused on the Right Ventricle)



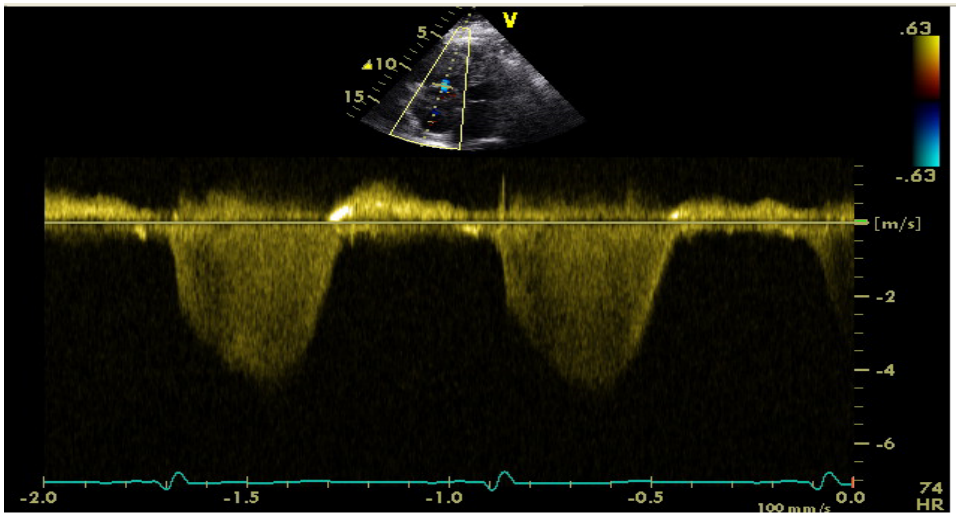
- The right ventricular length is maximized and the right ventricular apex is clearly visualized. The entire RV endocardium must be within the sector scan in both end-diastole and end-systole.

In addition to 2D imaging, the following Doppler examinations will be required from apical 4-chamber view focused on the right ventricle:

C.2.i. Color Doppler of tricuspid inflow

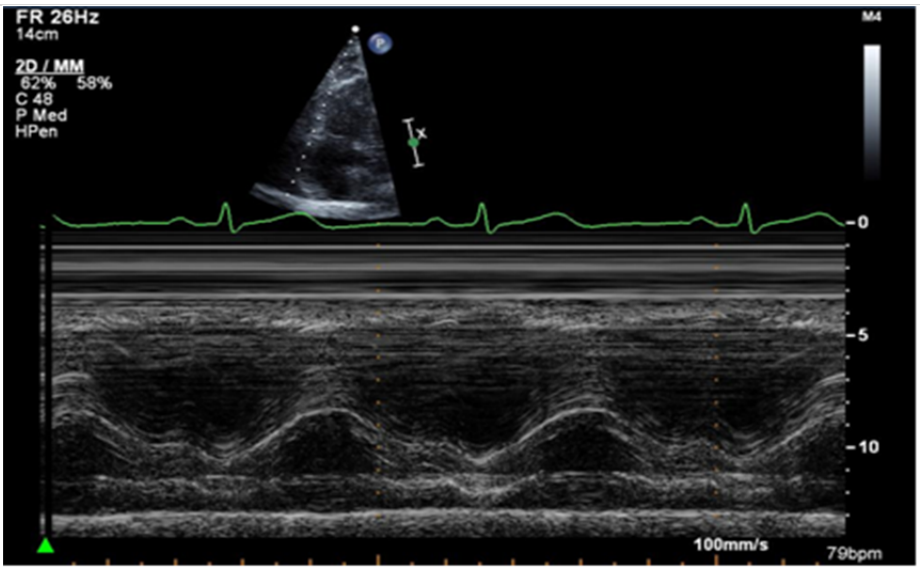


C.2.ii. CW Doppler of Tricuspid Regurgitation



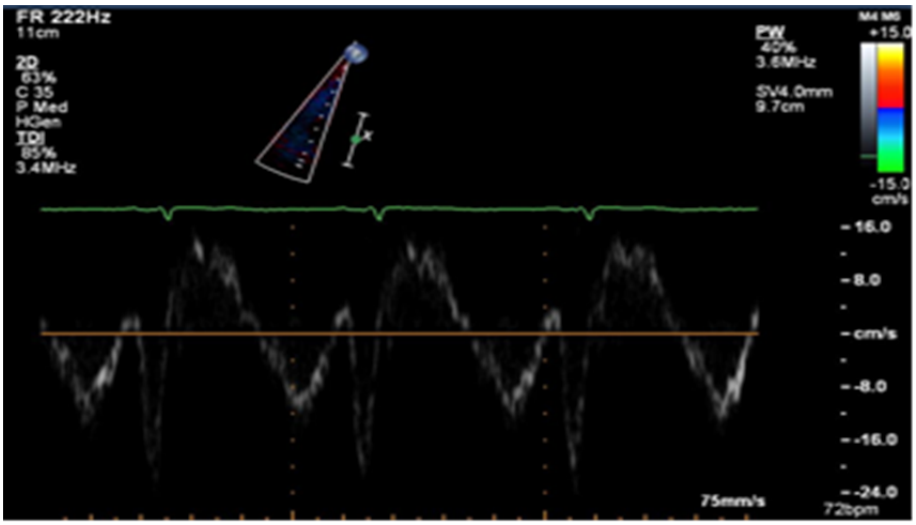
- Position the interrogation line down the right ventricle and atrium as parallel to tricuspid regurgitant flow as possible. Adjust the baseline and scale to capture the peak TR velocity. Record at least 3 (10 for subjects in atrial fibrillation) full representative systoles at sweep speed of 100 mm/sec.

C.2.iii. M-mode of the anterior tricuspid annulus



- Adjust apical 4-chamber image so M-mode cursor is aligned parallel to the anterior tricuspid annulus. Acquire M-mode images at a sweep speed of 100 mm/sec. Perform all recordings during apnea.

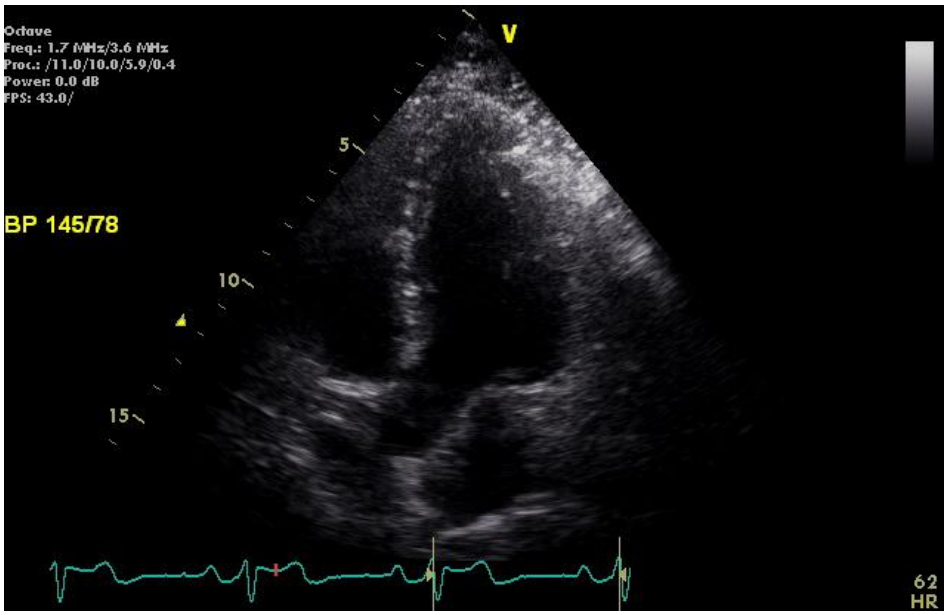
C.2. v. Tissue Doppler imaging at the lateral tricuspid annulus



- Decrease image depth to include the RV and a small part of the RA (optimal depth approximately 16 cm) and optimize the 2D image for the RV, focusing on the tricuspid annular region.
- Adjust the image to orient the motion of the anterior tricuspid annulus parallel to the cursor. Both gains and filter settings should be set low to obtain the best images.
- Initiate 2D color DTI and position the sample volume on the ventricular side of the lateral tricuspid annulus at the junction of the RV wall with the tricuspid annulus: the myocardium should stay within the sample volume for as much of the cardiac cycle as possible.

- Switch to PW spectral DTI and set the scale to 20 cm/sec or less with a sweep speed of 100 mm/sec. Once a clear pattern is obtained, record at least 3 beats during quiet respiration (or preferably during breath holding at end-expiration).

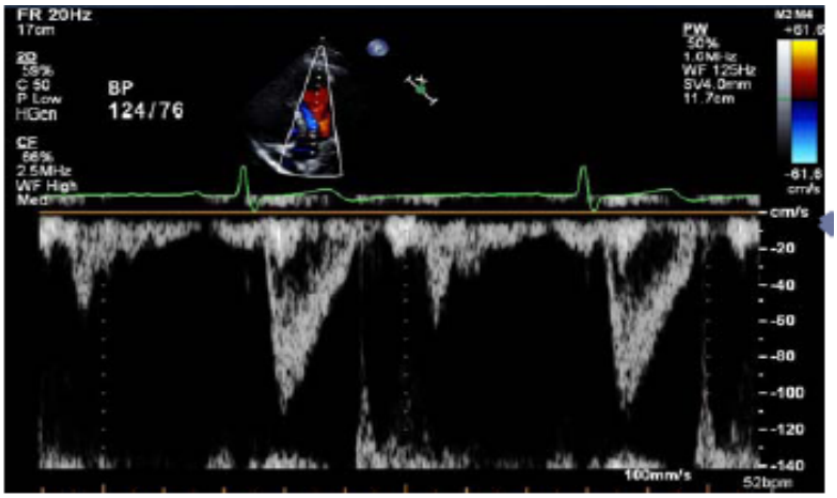
C.3. Apical 5-Chamber View

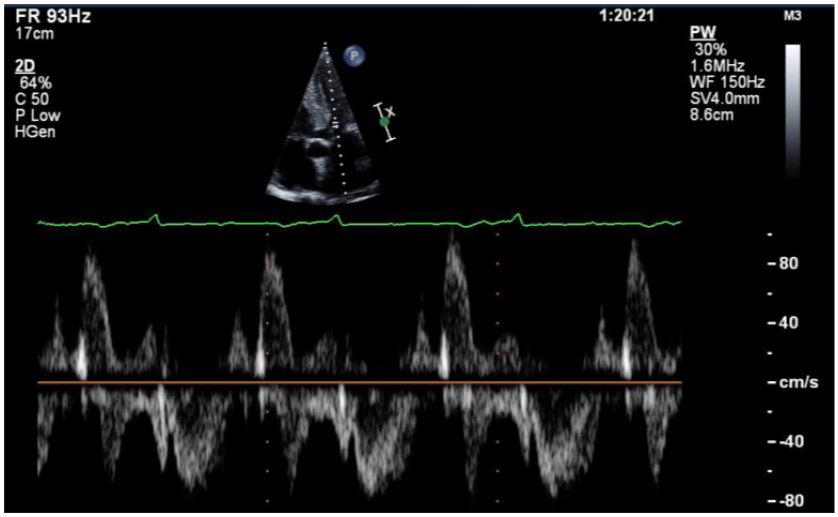


In the ideal echocardiographic "window" for the Apical 5-Chamber View:

- Angle transducer anteriorly to visualize LVOT, Aortic root and aortic valve in widest excursion. Maximize LVOT length, making sure not to truncate or foreshorten the true long axis of the LVOT. LV endocardium should be clearly visible.
- Place CW Doppler in LVOT and obtain flow with highest velocity and valve clicks. Switch CW to PW. Place PW sample volume in LVOT approx. 0.5 cm from the aortic valve.

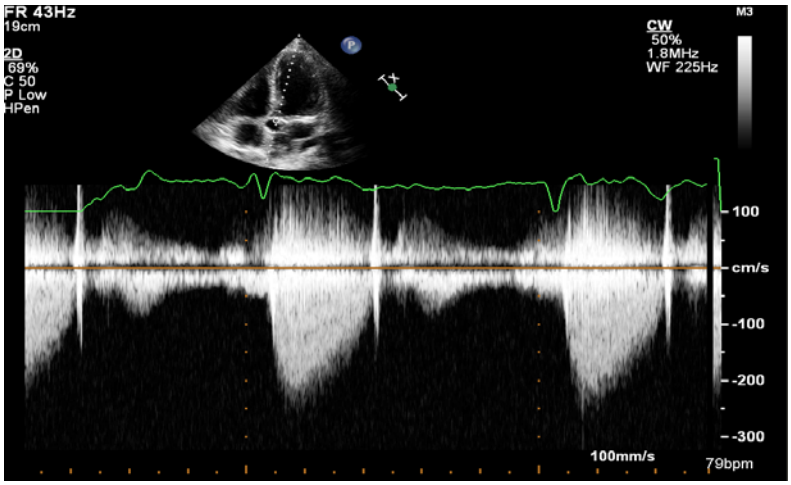
C.3.i. Pulsed wave Doppler at the left ventricular outflow tract



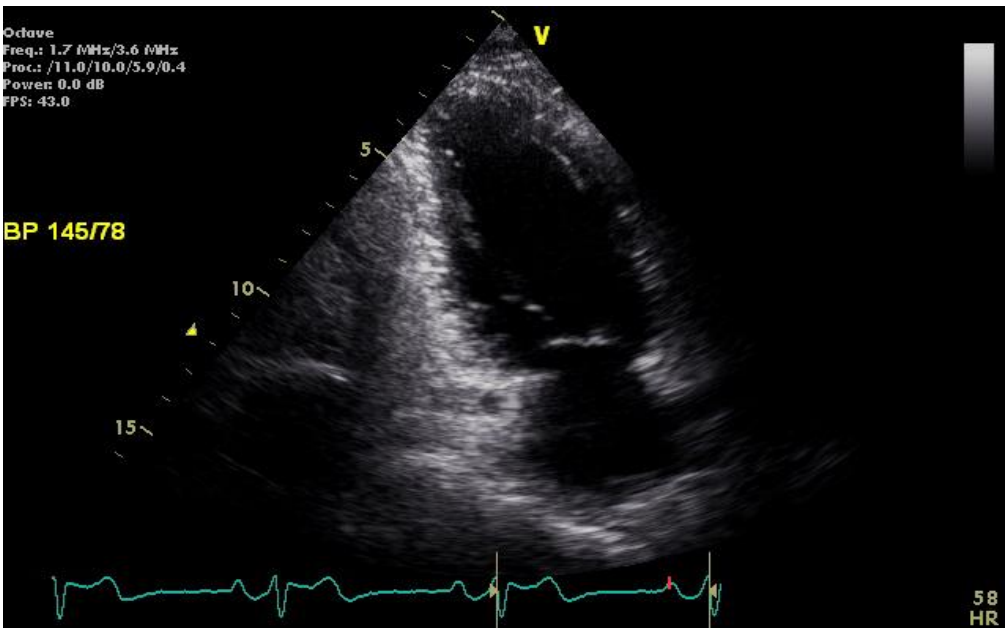


Record the isovolumic relaxation time (IVRT), defined as the interval from the closure of the aortic valve to the opening of the mitral valve. IVRT will be obtained with PW Doppler, the sample volume placed between the LVOTand the mitral inflow regions of the LV but in proximity to the anterior mitral valve leaflet along the mitral-aortic intervalvular fibrosa.

C.3.ii. Continuous wave Doppler across the aortic valve



C.4. Apical 2-Chamber View



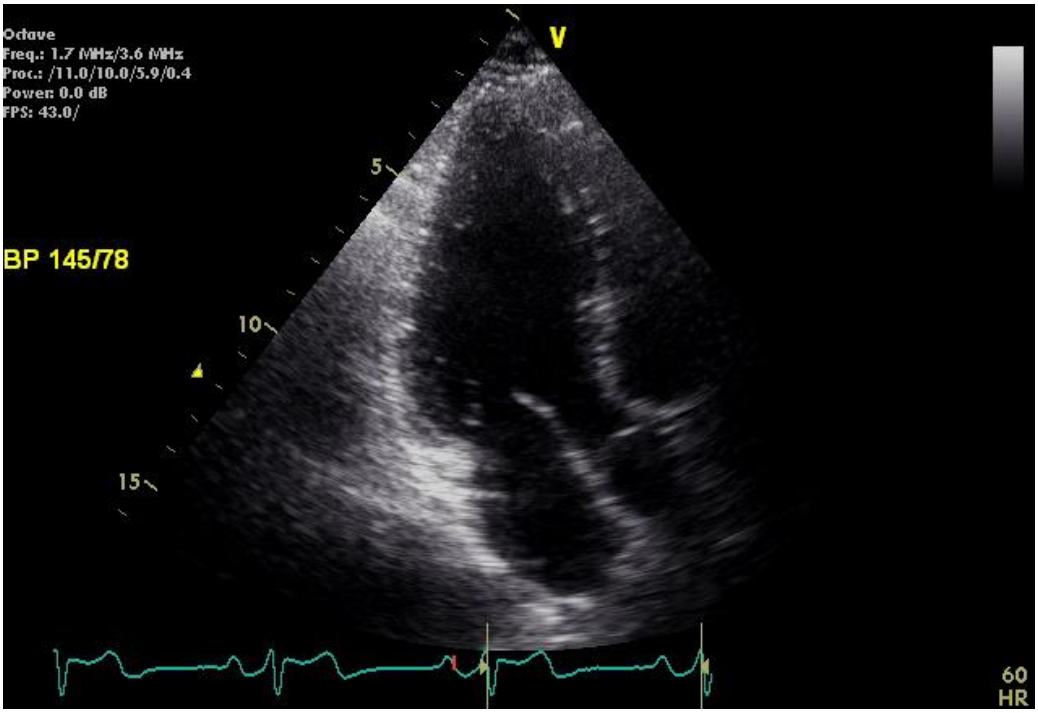
Obtain the apical two-chamber view, rotating the transducer at the apex 90° counterclockwise, so that the scan plane transects both the anterior and inferior LV walls; neither the RV nor the LV outflow tract should be visualized. Visualization of both anterior and inferior wall endocardium will be essential to accurately calculate left ventricular volume by Simpson's formula.

- Obtain 1 clip optimizing visualization of the left ventricle during systole and diastole. Obtain a second clip optimizing visualization of the left atrium throughout systole and diastole.

In the ideal echocardiographic "window" for the Apical 2-Chamber View:

- Maximize LV length and be careful not truncate the true long axis.
- The scan plane transects the anterior and inferior LV walls, with neither the RV nor the LV outflow tract visualized.
- The most difficult areas in which to visualize the endocardium are usually the anterior LV wall and the apex; pay particular attention to these walls**.**
- Special attention should be paid to properly aligning the image and capturing the left atrium in full. Avoid any foreshortening of the chamber.

C.5. Apical Three Chamber View:



Apical Three Chamber

- Obtain a 2D image, including the entire LA and LV and mitral valve
- Use color Doppler to display the mitral regurgitant jet.
- 1 clip Color AV (CW only if not acquired in 5-chamber view)

IV. Field Center Sonographer Training and Certification

Sonographer training and certification is a multilayer process and includes the following components:

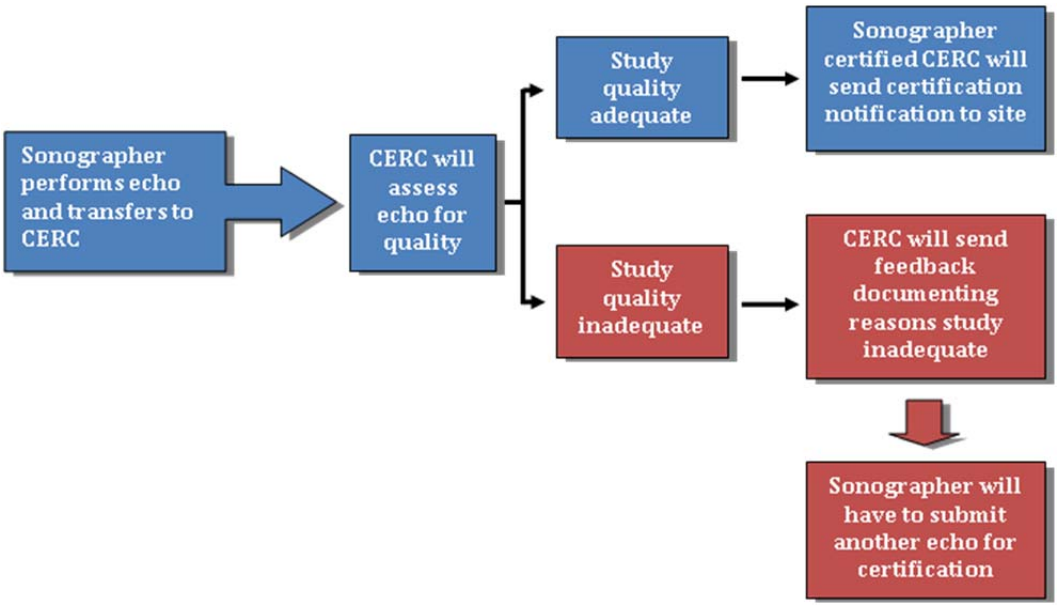
Live training session – A centralized training session will be held for all Field Center sonographers who will be performing echocardiograms for the ECHO-SOL. Training will consist of a didactic session, reviewing the exam protocol, machine presets, required views, image acquisition/optimization tips, and mechanisms for the Field Center staff to contact Cardiac Echo Reading Center (CERC) staff regarding technical questions or issues. A hands-on session will then be led by the CERC sonographer, initially demonstrating a full study exam on a model patient, then allowing each sonographer to perform the full exam under direct supervision. A presentation and demonstration of the process for transmitting studies to the CERC will be made.

Reference materials at the Field Centers – Prior to the ECHO-SOL recruitment period, the Reading Center will provide each site the **Field Center Manual of Operations** containing a detailed review of the imaging protocol (with illustrations) for Field Center sonographers, the timeline for echocardiography digital image acquisition, digital image capture instructions, instructions for image transmission, answers to frequently asked questions, technical tips for image optimization, and Core Lab contact information. The manual will include sample transmittal forms used to document key subject ID information, the name and contact information of the sonographer responsible for the study.

Sonographer Certification – The purpose of certification is to ensure consistency in how echocardiograms are performed study-wide and to ensure performance of the highest quality echocardiograms. Prior to site enrollment, any sonographer who will be performing study echocardiograms must first submit two certification studies performed in accordance with the protocol described in this manual and transferred electronically to the CERC for review and certification.

Test case review will be the final step prior to certification of sonographers and consists of a comprehensive itemized direct written feedback using a 40-item checklist of image quality components **(Section XV)**. A PDF of the feedback form will be e-mailed to the site within 48-72 hours of the receipt of the Certification Echo. Studies will be scrutinized for adherence to protocol, acquisition of all required views, and image quality and suggestions from the technical project manager will be provided for each study submitted. This is intended to address any individual equipment or operator dependent problems that may arise. Sonographers will have the opportunity to re-submit a sample protocol study should the initial submission be inadequate. Following submission of an adequate sample study, the sonographer will be officially certified and will receive feedback documenting this.

Sonographer Certification Process



New Field Center sonographers starting during the study period will be required undergo the certification process outlined above by submitting 2 sample protocol studies in order to demonstrate the ability to perform a technically adequate protocol study and the knowledge to successfully transmit this data to the CERC.

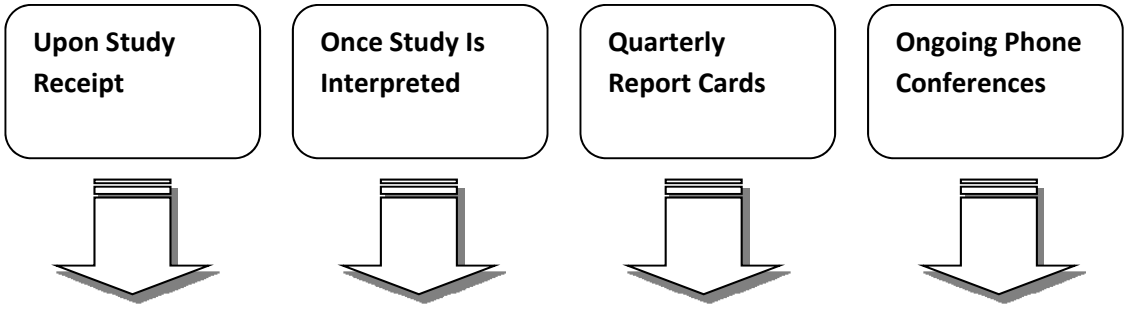
Monitoring and feedback – During the ECHO-SOL recruitment period, CERC technical staff will continuously monitor the adequacy and quality of all studies received.

Rapid review (same day of study receipt). For each study received, the core lab will review each study using a detailed 12-item checklist **(Section XV)** for completeness of the imaging protocol and study quality. Immediate feedback (both positive and negative) will be provided to each site via e-mail or phone discussion within 72-96 hours after the echocardiogram is received by the lab. If a study is deemed to be insufficiently performed, additional images will be requested whenever feasible.

Every time a study is interpreted, a 40-item detailed checklist (Section XV) will be filled out to provide detailed assessment of study adequacy. Within 2-4 weeks of study receipt detailed feedback, utilizing the itemized list regarding study adequacy and deficits, is sent to the Field Center via e-mail or phone. Study quality is judged by ability to visualize endocardium, adequacy of ECG gating, clarity of Doppler signals, adequacy of depth and scale optimization of images.

Report cards of each site's adherence to checklist items will be sent out after every 12 cases for the first six months and then on a quarterly basis. These will provide each site a picture of where that site falls relative to the other sites. It is expected that each site will have 90% adherence to the checklist items. A pattern of inadequate or poor quality studies will prompt directed discussion by CERC staff with the Field Center PI and/or sonographer and, possibly, retraining.

Ongoing phone conferences will be held bi-weekly for the first six months and then on a monthly basis. These conferences facilitate communication between the core lab and each study site. Each site PI, site echocardiographer collaborators, site sonographer, and study coordinator will attend. These phone conferences will be opportunities to address sites deficiencies, review protocols and offer technical assistance as needed.



- A Rapid review
- Using a 12-item checklist
- Immediate Feedback

- Within 2-4 weeks
- Using a 40-item checklist
- Study adequacy and deficits are judged

- Based on 40-item checklist
- 90% adherence is expected
- Shown relative to other sites

- Standing call
- Attended by site PI, cardiologist, site sonographer, study coordinator

V. Test Case Instructions

During the study set up period and at different points prior to initiating study enrollment, the Cardiovascular Core Laboratory at Wake Forest School of Medicine may request that you submit a "test case":

1. The "test case" can be performed on a subject of your choice (any volunteer, staff member, or a consenting patient). Your site coordinator will receive an e-mail notification that we have received, reviewed, and approved (or rejected) your test case.
    - You will not receive an official interpretation.
    - Recording media and transmittal sheets will be retained by the Core laboratory and are for training purposes only.

2. The purpose of the test case is to ensure that:
    - The required study elements can be obtained.
    - The study is recorded in a format that can be read and analyzed by the core lab.

3. If the "test case" reveals problems, we will work with you to correct any deficiencies.
    - Training will typically be by e-mail and/or phone.

4. Approval of two "test cases" is needed in order to be considered "certified" and begin to enroll participants.

5. Additional test cases may be requested in the future.
    - This will be at the discretion of the Core Laboratory and could be requested, for example, if your site has not performed the protocol for more than 6 months.

6. If you have any questions, the best way to reach us is by e-mail.
    - Our e-mail address is echo-sol@wakehealth.edu.
    - If necessary you may call us at 1-336-713-1648.
    - We are located in Winston-Salem, NC in the EST zone.

V. CERC Data Management Processes

All echocardiographic studies will be transferred from Field Centers to the Reading Center electronically using a secure web-based application pre-configured on a CERC provided computer. Field Centers will be notified via e-mail or phone upon successful receipt of the submitted studies.

Echo Data Transfer
A study specific e-mail account is established (echosol@wakehealth.edu) for the Field site Imaging staff to correspond with the Echo Core Lab and coordinate transfer of all echocardiographic data. Each study site will notify the core lab when study images are sent. The core lab will send return e-mails when the study is received and the initial review for image adequacy is completed.

The Echo Core Lab will work with each field Imaging site to configure networking as appropriate to their local needs to establish a two-way DICOM association. A password-authorized pre-configured computer system with DICOM send parameters will be provided to the field sites for routing the echo digital images to our server at Wake Forest University Health Sciences (WFUHS). DICOM image transfers will require two-way authorization and configuration and are conducted through an encrypted secure https protocol. While the data is transferring electronically it is encrypted using industry standard encryption methods. DICOM images are sent to WFUHS using the Commands Transfer Protocol (CTP) running on the pre-configured Windows computer. The CTP Field Center program forwards the encrypted images to the CTP DICOM receiver at WFUHS, then the images are forwarded to a secure DICOM PACS server for long-term storage and archiving.

The PACS server will also be used for permanent archival of all echocardiographic studies facilitating review and comparison of studies. If necessary, hard copies can be burned to CD's for storage from the PACS server files.

Use of DICOM transfers between the field sites and reading centers eliminates the handling of media (CD's and optical disks), because media are easily copied, lost or stolen. This means of transferring image data was chosen because it provides instant data transfer and is more efficient and cost-effective than mailing/courier service. This should improve turnaround time for result reporting with the field centers. Echo Core Lab Philips *Xcelera*® workstations are configured to query/retrieve the DICOM PACS server to quickly and easily access the digital echo images in their original preserved DICOM format with all calibrations remaining intact. Only de-identified data will be transferred according to HIPAA guidelines. The data will be protected by a user name and password protocol.

For analysis of established parameters of cardiac structure and function, the CERC utilizes commercially available and validated *Xcelera*® analysis software which allows for standard echocardiographic analysis from digital (DICOM) echocardiograms. The software is capable of making all standard echocardiographic measures, including ventricular volumes and LVEF via modified Simpson's method. All 2D speckle-tracking measurements will be performed using the TomTec® software. 2D Cardiac

Performance Analysis (2D CPA) is a vendor independent solution dedicated for strain, strain rate and velocity analysis based on speckle tracking. 2D strain data are extracted into a spreadsheet for the generation of time velocity and strain curves from apical or parasternal views.

All analyses will be formally over-read by the ECHO-SOL PI or co-I's. Over-readers will be assessing study echocardiograms for critical abnormalities that may require clinical attention and impact study subject care and for standard clinically reportable measurements that will be used to generate clinical alerts. Over-readers will not be re-measuring values, unless necessary, but reviewing both images and measurements to ensure appropriateness of reported measures.

Echo data tracking (within core lab)

Transfer forms will be used to record participant scans completed and transferred to the Core Lab. Discrepancies will be reconciled between the core lab and each Field Imaging Center promptly, and both centers will maintain electronic copies of transfer forms throughout the study.

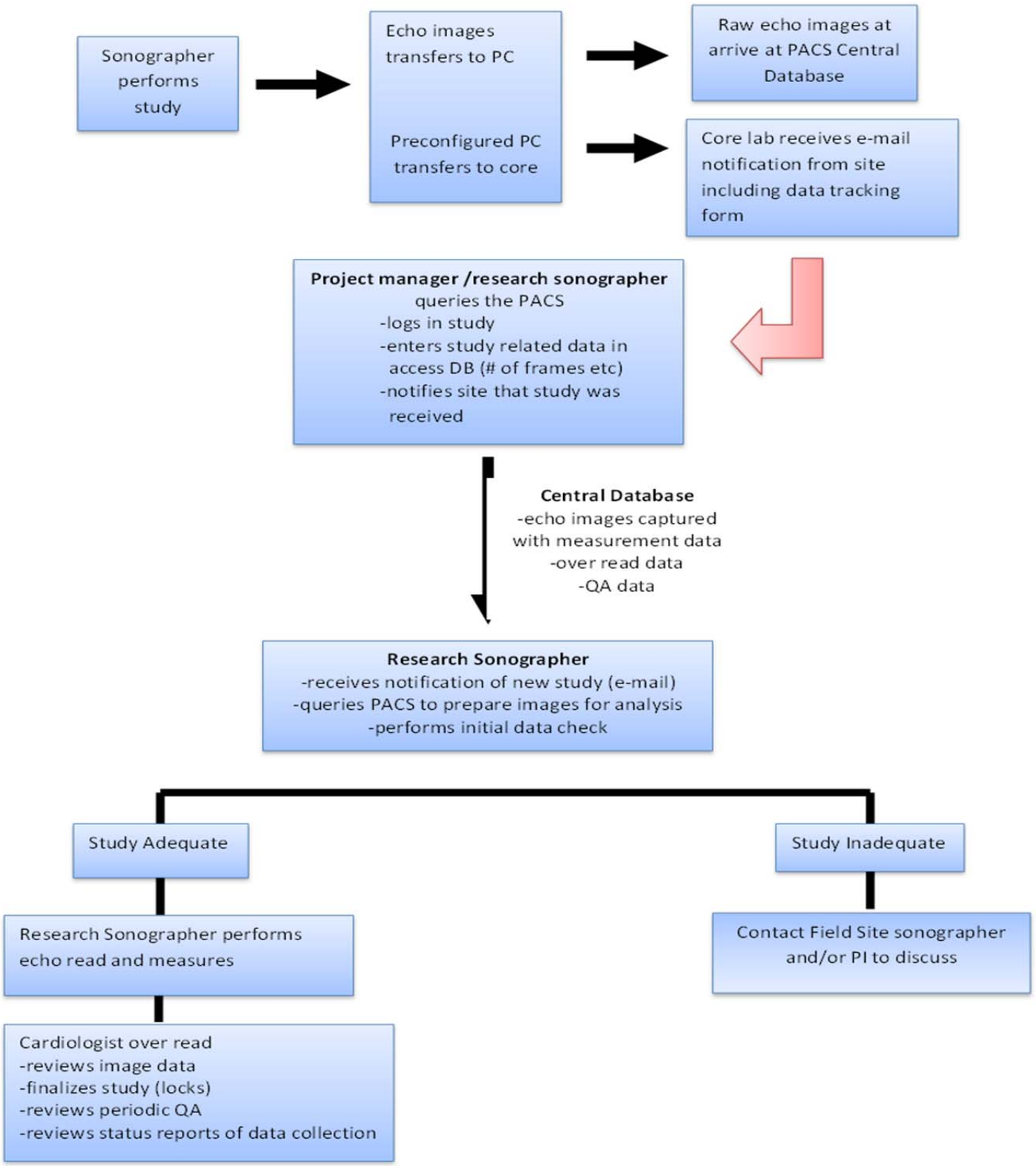
Email notification from each site including a completed Data Tracking form is key. Upon receipt of email notification, the CERC staff will query the PACS to assure that the raw echo images have arrived. Upon receipt, studies will be logged into a custom-made RedCap database. The database will be used to track all steps in the study accession and review process throughout the CERC (including HCHS-SOL ID, study date performed, number of frames, sonographer initials, date of receipt, date of site notification, site contact, date of initial review, reader started date / completed date, over read date, comments, clinical alerts yes / no, alerts sent date, QC reader assigned, 2D speckle started date / completed date).

Echocardiography measure data will be electronically exported from the Philips *Xcelera*® workstations onto a custom-made spreadsheet whose upkeep will be the responsibility of the Study Coordinator. These data will undergo a preliminary check for any gross outliers, anomalies or problems before being uploaded onto a SAS database by our local analyst. Data will be periodically electronically transmitted to the Data Coordinating Center where they will be further cleaned, uploaded and collated into the main HCHS-SOL database.

Long term storage of materials

All Core Lab data files are saved and backed up regularly. Digital images will be stored in a segregated section of the DICOM PACS as well as on the file server used for cardiac ultrasound image archival both which is backed up daily. All hard copy digital materials (CD's, DVD's) are duplicated and stored in secured storage space. Materials specific to this study will be maintained for at least seven years or longer as deemed necessary.

VII. ECHO-SOL Study Acquisition Timeline / Algorithm



VIII. Echo Reading Center Measurements

We will require duplicates measurement from each cardiac cycle.

1. Assessment of Global Left Ventricular Size and Function

From the parasternal long or short axis view/M-Mode below the tips of the mitral valve, the end-diastolic dimensions as well as the interventricular septum thickness and posterior wall thickness of the left ventricle will be measured according to the recommendations of the American Society of Echocardiography.

From the 4-chamber and 2-chamber apical views, the endocardial borders will be traced in both end-diastole and end-systole to obtain left ventricular areas.

Left ventricular volumes will be obtained utilizing the Simpson's rule algorithm and average of 4-chamber and 2-chamber single plane volumes.

Parasternal long axis view:

- End-diastolic left ventricular diameter (cm) [LVIDd]
- End-systolic left ventricular diameter (cm) [LVIDs]
- Interventricular septum thickness [ISWT]
- Posterior wall thickness [PWT]
- Left ventricular overflow tract [LVOT] diameter

Apical 4-Chamber View

- End diastolic volume (ml) [EDV]
- End systolic volume (ml) [ESV]

Apical 2-Chamber View

- End diastolic volume (ml) [EDV]
- End systolic volume (ml) [ESV]

<u>Derived Measurements</u>

- Ejection fraction (%) = 100 *(LVEDV – LVESV)/LVEDV
- LV mass (g) = $0.8*\{1.04*[(LVIDd+IVSTd+PWTd)^3 - (LVIDd)^3]\}+0.6$
- LVOT area = 2 pi $(r)^2$

2. Assessment of Left Ventricular Diastolic function:

The peak velocities of the early rapid filling wave (E wave), and the peak velocity of the late filling wave (A wave) will be measured. The deceleration time of the E wave will be measured as the interval from the peak E wave to its extrapolation to the baseline.

Mitral annular velocities (for tissue Doppler imaging) will be recorded in systole and diastole at the lateral and septal annulus in 4-chamber view. Early diastolic myocardial velocity (E') and late diastolic myocardial velocity (A') will be measured at the lateral and septal annulus. Measure TDI E' and A' at the apex of the triangle.

Interrogation of Pulmonary Venous (PV) Flow involves placement of a Doppler sample volume 1-2 cm into the lumen of the right upper PV and recording systolic (S) and diastolic (D) inflow velocities. PV atrial reversal (Ar) velocity >35cm/s or the time difference between Ar duration and mitral A-wave duration (Ar - A) > 30ms, both serve as indicators of elevated LV end-diastolic pressures.

Apical 4 Chamber View

Mitral Inflow Doppler at the tips of the mitral leaflet

- Peak E wave velocity (cm/sec)
- Peak A wave velocity (cm/sec)
- Deceleration time (msec)

Tissue Doppler Imaging

- Lateral early diastolic myocardial velocity ($E'_{lateral}$)
- Lateral late diastolic myocardial velocity ($A'_{lateral}$)
- Septal early diastolic myocardial velocity (E'_{septal})
- Septal late diastolic myocardial velocity (A'_{septal})

Pulmonary Vein Doppler (Pulse Wave)

- Systolic velocity (cm/sec)
- Diastolic velocity (cm/sec)
- Atrial reversal velocity (cm/sec)

<u>Derived Variables:</u>

1. E/A ratio

2. E/ $E'_{lateral}$ ratio
3. E/ E'_{septal} ratio
4. S/D ratio

Apical 5 chamber view

- Isovolumetric relaxation time (msec)
- LVOT VTI for Stroke Volume

3. Assessment of Left Atrial Size

From the parasternal long axis view, the maximal left atrial diameter will be measured according to the recommendations of the American Society of Echocardiography.

From the apical 4- and 2-chamber end-systolic views, left atrial endocardial borders will be traced allowing for the measurement of left atrial volumes by modified Simpson's rule. The inferior border is represented by the plane of the mitral annulus. The pulmonary veins and left atrial appendage are excluded from the left atrial tracing.

Parasternal long axis view

- Maximal left atrial diameter (cm)

Apical 4 Chamber View

- 4-Chamber left atrial volume at mitral valve opening (ml)

Apical 2 Chamber View

- 2-Chamber left atrial volume at mitral valve opening (ml)

Derived Variable:

- LA volume index (ml/m^2) = LA volume/BSA

4. Assessment of Global Right Ventricular Function

From the apical 4-chamber view focused on the right ventricle, the endocardial borders will be traced in both end-diastole and end-systole to obtain right ventricular areas.

From the apical 4-chamber view TDI of the right ventricle lateral tricuspid annulus and peak systolic velocity (S'_{TA}) will be measured.

Peak tricuspid regurgitation velocity will be measured using continuous wave Doppler through the tricuspid valve.

Parasternal short axis view

- RVOT VTI for Stroke Volume
- PV annular diameter

Tricuspid Regurgitation Doppler (Continuous Wave)

- Peak tricuspid regurgitation velocity (m/sec)

Apical 4 Chamber View

- RV end diastolic area (cm2) [RVEDA]
- RV end systolic area (cm2) [RVESA]

Tissue Doppler Imaging

- Lateral systolic myocardial velocity (S'_{TA})

Derived Measurements

- RV fractional area change (unitless) = (RVEDA – RVESA) / RVEDA

5. Assessment of Left Ventricular Deformation

From the 4-chamber and 2-chamber apical views, the endocardial and epicardial borders will be traced for measurement of longitudinal and transverse strain utilizing semi-automatic border tracking using the TomTec® software.

From the parasternal short axis at the level of the mitral valve, mid-papillary muscle, and apex, endocardial and epicardial borders will be traced for measurement of radial strain, circumferential strain, and rotation.

Parasternal short axis view (mitral level, mid-papillary level, apex):

- Radial strain
- Time to peak radial strain (msec)
- Circumferential strain
- Time to peak radial strain (msec)

Apical 4 Chamber View

- Longitudinal strain
- Time to peak longitudinal strain (msec)
- Transverse strain
- Time to peak transverse strain (msec)

Apical 2 Chamber View

- Longitudinal strain
- Time to peak longitudinal strain (msec)
- Transverse strain
- Time to peak transverse strain (msec)

Derived Measurements

- Average peak longitudinal strain
- Standard deviation in time to peak longitudinal strain (msec)
- Average peak transverse strain
- Standard deviation in time to peak transverse strain (msec)
- Average peak radial strain
- Standard deviation in time to peak radial strain (msec)
- Average peak circumferential strain
- Standard deviation in time to peak circumferential strain (msec)
- Peak left ventricular torsion

IX. Reporting of Findings to Field Centers

The CERC will be reviewing echocardiograms performed in ECHO-SOL. These studies are performed to obtain quantitative measures of cardiac structure and function in a Hispanic community-based cohort. There are several limitations to the studies: the studies are not clinically indicated and a comprehensive ASE recommended transthoracic study is not being performed. Additionally, the CERC will be reviewing studies in the absence of any clinical information. **For these reasons, CERC adjudication of ECHO-SOL echocardiograms will not and cannot be equivalent to a clinically acquired and interpreted echocardiogram.**

Findings to be reported

For routine studies, we will provide a qualitative summary letter echo report within 6-8 weeks of study receipt for the participant. This report will be e-mailed to the site PI and site study coordinator as a PDF file. The sites will give this summary letter to the participant. Qualitative analysis reporting of findings is provided purely as a courtesy to ECHO-SOL participants. It is not meant to supplement a full clinical echocardiographic assessment. Quantitative assessment of right ventricular function is not standard in clinical practice and parameters of RV function (RV fractional area change and lateral tricuspid annular S' by tissue Doppler) will not be reported. Similarly, pulmonary vascular resistance, 2D speckle tracking measures of strain and strain rate determined by echocardiography are not clinically measured and will not be reported.

It is important to note that routine studies may not undergo full final analysis for 6-8 weeks. Thus it is imperative that the site sonographers at the time of echocardiogram performance note any gross critical and non-critical abnormalities to the site PI and cardiologist as outlined below.

Abnormal BP protocol

If a systolic blood pressure of 180 mmHg or above, or a diastolic of 110 mmHg or above is detected (by two measurements 5 minutes apart at the time of the echocardiogram), the site PI and/or site cardiologist needs to be notified. If the participant is asymptomatic, the participant will complete their echocardiographic exam. The participant's blood pressure will be checked again by the site PI or designated surrogate. If elevation above 180 mmHg is confirmed, then outpatient or ER evaluation will be offered to the participant.

Graded alert system

Critical Abnormalities

For potentially serious abnormalities that may require urgent/emergent medical evaluation, the Field Center sonographers will be instructed and expected as first line of screening for critical and non-critical abnormalities, to report any of the following findings to the Field Center site PI **immediately** at the time of echocardiogram performance and to the Core Lab at the time of echocardiogram study transmission. These include, but are not limited to:

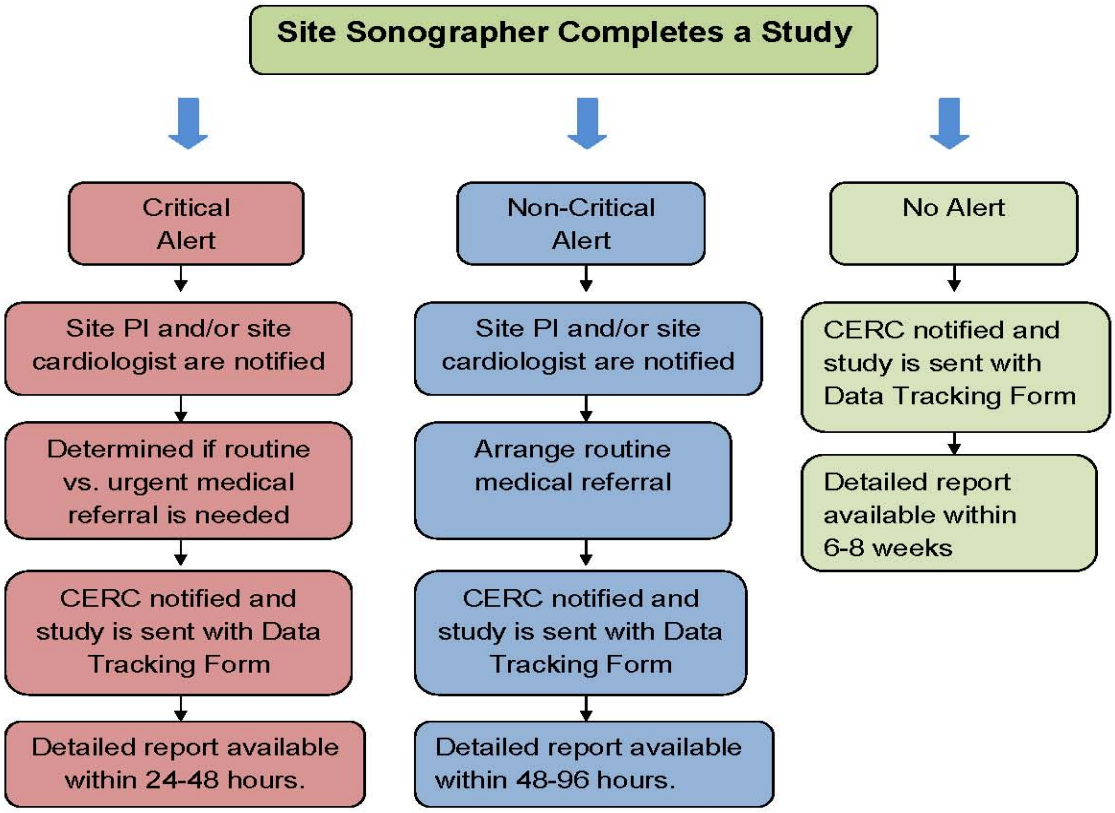
Critical Abnormalities
1. Tamponade
2. Aortic dissection
3. Thrombosed or frankly dysfunctional prosthetic valve
4. Pseudoaneurysm
5. Intracardiac abscess or obvious vegetation
6. Intracardiac thrombus or mass

Non-Critical abnormalities
1. Moderate or greater mitral regurgitation
2. Moderate or greater mitral stenosis
3. Moderate or greater obstructive lesions of left ventricular outflow, including aortic stenosis and dynamic left ventricular outflow tract obstruction
4. Moderate or greater aortic regurgitation
5. Moderate or greater aortic stenosis
6. Moderate to severe pulmonary hypertension
7. Severe left or right ventricular enlargement.
8. Low ejection fraction or wall motion abnormality

For critical and non-critical abnormalities noted above, a **critical** alert will be generated by the site sonographer. The Field Center PI or designated surrogate will timely review all study alerts, verify the findings and the alert classification, and determine whether urgent or routine referral is needed and how results should be communicated to participants and / or their personal physicians. The Field Center will alert the CERC via phone or e-mail and an expedited analysis of the study will be performed. A more detailed echocardiographic report will be generated and e-mailed to the site PI as a PDF file within 24 hours for critical abnormalities and within 48 hours for non-critical abnormalities.

In the event that cardiac abnormalities are missed at the local site level, the CERC will serve as the second line of screening for critical and non-critical abnormlaities. **On the same day of study receipt**, the CERC research sonographer **performs a rapid review** of each submitted study for completeness and adequacy of images as well as assessing for gross abnormalities that may require clinical attention. Any abnormal findings will be verified by the CERC PI. Abnormal studies will be flagged at the CERC, and the site study coordinator and site PI or designated surrogate will be notified immediately. Locally licensed cardiologists at each of the Field Centers are responsible for reviewing and verifying the findings and alerts and making determinations about communication of the reports to participants' local physicians. A more detailed quantitative analysis reporting of findings will be generated within 48-96 hours of study receipt and be given to the Field Center site PI and site ECHO-SOL staff who will then, at the participant's consent, share the finding with the participants' local physicians, who may then be solely responsible for determining any appropriate follow-up or clinical care.

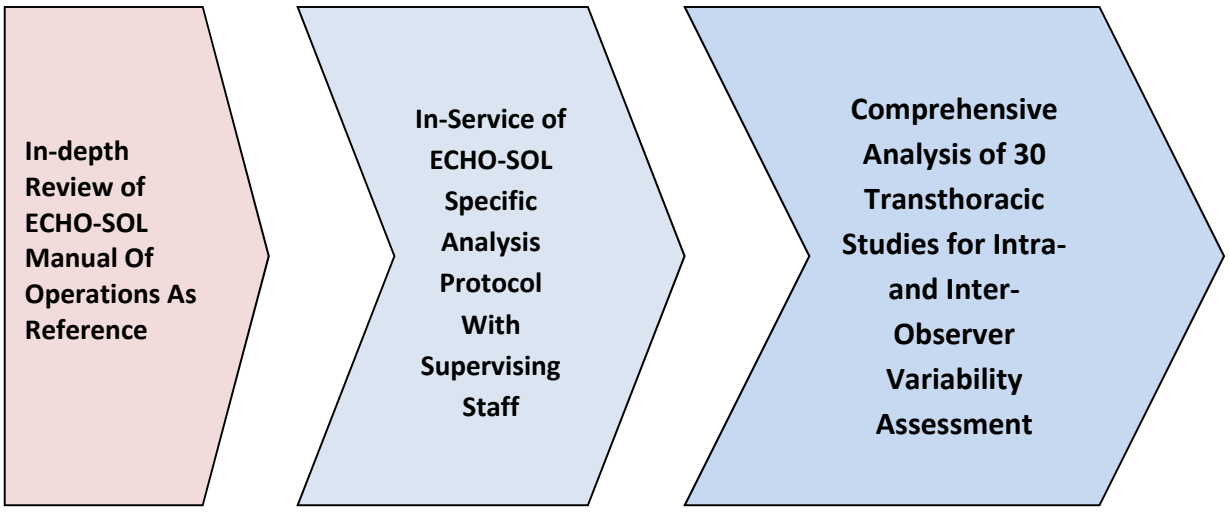
Clinical Alert Flowchart



X. CERC Echo Research Sonographer Training and Certification

Prior to enrollment, Reading Center Research Sonographer assessment for Intra- and Interobserver variability will consist of a comprehensive analysis on 30 transthoracic studies. Technical staff involved in the ECHO-SOL will undergo an additional in-service to thoroughly familiarize them with the analysis protocol. Duplicate blind re-reads of approximately 15 studies originally analyzed by the same technician for intra-observer reproducibility and 15 studies originally analyzed by an experienced echocardiographer for inter-observer reproducibility.

Technical staff involved in the 2D speckle-tracking strain and strain rate analysis will have to demonstrate acceptable intra- and inter-observer reproducibility of average longitudinal and circumferential strain measures.



XI. Quality Assurance Plan

Quality Assurance (QA) of Field Imaging Centers

QA involves activities that take place prior to data collection. QA activities in this study include development of the protocol for data acquisition and interpretation of the echocardiograms, adaptation of data entry and data management systems, training and certification of sonographers and pilot testing of the data collection procedures.

1. For consistency, one sonographer per site will be chosen as a dedicated sonographer for this study. Only certified technicians with national Registered Diagnostic Cardiac Sonographer (RDCS) credentialing will be assigned. RDCS sonographer credentialing requires that the clear, established, professional standards for echocardiographic data acquisition are adhered to. All chosen sonographers will be expected to adhere to this standard. The Core Lab will provide ongoing support for study sites as it pertains to the acquisition of echocardiographic data.

2. Centralized training site will be provided. The four chosen sonographers will visit Wake Forest University where the echocardiography imaging protocol will be reviewed in detail. All sonographers will perform the echocardiography imaging protocol under the supervision of the ECHO-SOL PI and lead sonographer by having volunteers scanned twice by the same and by different technicians. These will provide a forum for interactive case review, discussion of technical issues related to image acquisition, and resolution of any ambiguities relative to the protocol.

3. Prior to site enrollment, each sonographer will send test cases from their respective Field Imaging Center. Once the test cases are reviewed demonstrating the ability of the sonographer to properly acquire all of the echocardiographic information required for the study, the sonographer will be certified. Test case review consists of a detailed checklist of image quality components**.** The interaction involved in the test case approval process helps establish effective lines of communication and a degree of personal connection between Core Lab and study sites personnel.

Quality Control (QC) of Field Imaging Centers

QC consists of activities during and after data collection. Ongoing QC of individual site echocardiographic data acquisitions will be accomplished by detailed study by study feedback as well as ongoing quarterly review for performance monitoring of personnel at clinical sites.

1. **Rapid review (same day of study receipt)** of each submitted study will be performed for completeness and adequacy of images using a 12-item checklist **(Section XV).**
2. **Every time a study is interpreted, a detailed 40-item checklist (Section XV) will be filled out to provide detailed assessment of study adequacy.** It is expected that each site will have 90% adherence to the checklist items. Report cards will be sent out after every 12 cases for the first six months and then on a quarterly basis to each site to provide a picture of where that site falls relative to the other sites.

3. **Ongoing phone conferences** for the site PIs will be held biweekly for the first six months and then on a quarterly basis. Phone conferences for the study coordinator, study sonographer and site coordinators/sonographers will be held monthly. To facilitate communication between the core lab and each study site, we have identified experienced echocardiographer collaborators at each site. In addition, one-on-one discussions with Field site investigators will be held on an as needed basis. These phone conferences or one-on-one discussions will be opportunities to address sites with persistent deficiencies, review protocols and offer technical assistance as needed.
4. **Annual investigator meetings** will be carried out each year over the course of the proposed study. The site investigators and Field Imaging Site Directors will meet once a year at a national location corresponding with the American Heart Association or American College of Cardiology or American Society of Echocardiography Scientific Sessions.

QC of Echo Core Lab

Initial review of echocardiographic studies will be performed offline at the Echocardiography Core Lab by a registered sonographer who will prepare the studies to be over read and finalized by an experienced echocardiographer blinded to the subject's clinical and demographic characteristics. The latest release Philips *Xcelera*® review stations are available for use. These have excellent image quality and permit auto calibration for images acquired using the DICOM standard with most commercially available systems. Studies will be read at a capacity of 12-16 studies per week. The purpose of the Reading Center quality control procedures are to: (1) quantify intra-observer reproducibility, (2) quantify inter-observer reproducibility, and (3) quantify and mitigate temporal drift in echocardiographic analysis over the study period.

Intra- and interobserver variability – will be assessed on a **quarterly basis** throughout the duration of the study by use of intraclass correlation coefficients for the variables measured. A 5% random sample of each reader's images during the quarterly period will be reread by a second reader to assess for inter-observer variability. In addition, a 5% random sample of each reader's images will be reread by the same reader to assess for intra-observer variability. Variability testing will include equal representation of studies from each Field Center (anticipated 180 studies, 45 studies per Field Center). Re-reads will be performed in a blinded fashion to the original assessments. If significant variability is identified (>10% difference) for key echocardiographic measures (LVEDV (by method of discs); LVESV; LVEF; TDI E' velocity; LVM), the studies will be reviewed by all readers together to identify sources of error and reconcile differences.

Temporal drift – To assess for temporal drift for both established and 2D speckle-tracking measures, each reader will be required to perform blind re-reads on same set of 10 studies at 6 month intervals (total of 40 re-analyses per reader). Reproducibility of key measures will be assessed using the Bland-Altman method to compare repeated measures, with the coefficient of variation and bias reported.

Reporting of QA Assessments
Data regarding *intra- and inter-observer variability, temporal drift* for key echocardiographic measures will be reported to the Data Coordinating Center every 6 months. Reproducibility results will be reported primarily as the coefficient of variation, bias, and limits of agreement.

Summary of QA Plan:

Task	Pilot Phase	During Study Course	Reporting to coordinating center
CERC technician Intra-observer reproducibility	Duplicate blinded reads of 15 studies previously read by same technician	Every 3 months, repeat blinded reads of 5% of studies performed per technician during that period	Every 6 months
CERC technician Inter-observer	Duplicate blinded reads of 15 studies previously read by an experienced echocardiographer	Every 3 months, repeat blinded reads of 5% of studies performed per technician during that period	Every 6 months
CERC technician temporal drift		Duplicate blinded reads of the same 20 studies every 6 months	Every 6 months

XII. Echo-Sol Technical Tips

1) It is important that the parasternal long axis view displays the true long axis of the ventricle with the left ventricle lying horizontally on the image. It is unacceptable to record and off-axis view in which the apex “points up” on the screen. If this type of image is obtained try moving the transducer up an interspace or 2 or having the patient take a breath in. Sometimes having the patient move to a more lateral decubitus position will help as well.

2) If you are having difficulty locating the right upper pulmonary vein, lower the color scale and have the patient take a breath in.

3) For each view, the gain and compression should be optimized so that the best echocardiographic image of the endocardial borders is obtained. The selection of harmonics or fundamental frequency should depend upon which yields the best definition of structures.

4) All images should have a good quality ECG tracing on the screen and clear calibration markings on the imaging sector. For Doppler spectral tracings, the sweep speed should be at least 100 mm/sec and the scale and baseline should be adjusted to make sure that the entire Doppler envelope is visualized.

5) Time and velocity calibration markers must be present on the Doppler tracing. For spectral and color Doppler the appropriate gain level should be selected that detects flow without extraneous noise or extension of signal into adjoining tissue.

6) Because LV volumes from the apical views are an important end-point of the study, please try to avoid apical foreshortening. If the view appears to be foreshortened, please bring the transducer down one interspace and have the patient take a breath in. Sometimes this will bring out a better (not foreshortened) view.

7) Spectral Doppler (pulse wave and continuous wave) should be performed with the line of interrogation as parallel to flow as possible. Record Doppler at 100 mm/sec sweep speed or greater.

8) To facilitate your study acquisition, the sequence outlined here is similar to that used in many clinical labs. However, even if it deviates from your lab routine, the sequence of views must be followed as written. Within each view, the sequence may be changed as long as all elements are recorded.

9) Do not perform on-line measurements. Measurements will be performed at the Wake Forest School of Medicine Echo Core Laboratory.

10) Please do not hesitate to e-mail us at **echosol@wakehealth.edu** if you have any questions. Indicate a name, telephone number, and ideal call-back time (with your time zone) in case your question cannot be easily handled by e-mail. For urgent questions, you can also call **336-716-6498 or 917-658-0947**.

XIII. CERC STAFF DIRECTORY

Study Specific E-mail Account - (echosol@wakehealth.edu)

Study Coordinator
Sarah Langdon, MPH, CHES*
336-713-7613
slangdon@wakehealth.edu

Research Sonographer
Rupal Dadhania, RDCS*
rdadhani@wakehealth.com

Rhonda Blaine, Administrator*
336-713-0186 or
Email rblaine@wakehealth.edu

IT "hot line"**
Jerry Holbrook
(336) 713-5072 or
jeholbro@wakehealth.edu

Dr. Carlos Jose Rodriguez, PI
336-713-1648
Cell 917-658-0947
crodrigu@wakehealth.edu

*** Please try these first if you need assistance**

**** Do not use the echosol inbox for IT related issues. Please direct all IT questions directly to Jerry Holbrook preferably via e-mail.**

XIV. ECHO-SOL Cardiac Echo-Doppler FAQs

Q: ***What happens if a piece of data is missing or suboptimal? Will I receive feedback on the quality of my exams?***
A: If the Core Laboratory is unable to interpret the data due to a deficiency or poor quality, you, the PI and the site coordinator will be notified by e-mail. In some cases, the exam may need to be repeated if the study is non-diagnostic or if you have deviated from the standard protocol. In other cases, you may receive constructive feedback for use in future exams. We will also try to provide positive feedback on adequate high quality studies. Every quarter, in which you have performed studies, you will receive a "report card" as to the adequacy of key echo elements. This is intended to help you see what we see in terms of analyzable data. We are aiming for at least 90% analyzability.

Q: ***What should I do if there is an unexpected finding that, in my opinion should be conveyed to the patient's physician immediately to ensure optimal patient care (for example left ventricular thrombus, severe and not previously documented valve stenosis or regurgitation)?***
A: Patient care is paramount. Please notify attending physicians as per your usual lab protocol.

Q: ***What if I make a mistake on the form?***
A: Errors should be crossed out with a single line, initialed, and dated.

Q: ***What are the usual reasons for requiring a repeat exam?***
A: The most common problems include:
- Deviation from the protocol or an incomplete exam.
- Mislabeling the tape, data sheet, or image.
- Missing data on the data sheet.
- Poor image quality.

Q: ***When can I start with my first participant?***
A: You must be "certified" with your test case prior to proceeding with your first subject. Your site coordinator will be notified when you are certified.

XI. Forms

Data Tracking Form
ECHO-SOL Visit 2
Cardiac Echo Reading Center

An e-mailed or faxed copy of this tracking form must accompany each image sent to the Cardiac Echo Reading Center

1. HCHS-SOL ID:	
2. STUDY SITE	Choose an item.
3. Date of Echo Exam:	Click here to enter a date.
4. Clinical Alert Generated:	Choose an item.
5. If Yes to #4, describe the abnormality	Choose an item.

Physical Assessment		
6. Blood Pressure	(mm/Hg)	☐ Not Done
7. Height	(cm)	☐ Not Done
8. Weight	(kg)	☐ Not Done

My signature indicates that to the best of my knowledge all information entered on this form is correct.

	Click here to enter a date.
Sonographer's Initials	**Date** (mmm/dd/yyyy)

SITE FEEDBACK FORM
ECHO-SOL Visit 2 Cardiac Echo Reading Center

Study date: Date _____ **Date received by lab:** Date _____ **HCHS-SOL ID:** _____

Sonographer Initials: _____ **Internal Use:** _____

Criteria to Meet	Completed Correctly	Comments / Tech Tip
2D and Color Long Axis	☐Yes / ☐No / ☐NA	
2D and Color Short Axis	☐Yes / ☐No / ☐NA	
2D Four Chamber with good endocardial definition	☐Yes / ☐No / ☐NA	
Color AV and MV in Four Chamber	☐Yes / ☐No / ☐NA	
MV Inflow	☐Yes / ☐No / ☐NA	
Tissue Doppler	☐Yes / ☐No / ☐NA	
Pulmonary Vein Inflow	☐Yes / ☐No / ☐NA	
PW LVOT	☐Yes / ☐No / ☐NA	
Aortic Valve CW	☐Yes / ☐No / ☐NA	
2D Two Chamber with good endocardial definition	☐Yes / ☐No / ☐NA	
Color MV in 2 Chamber	☐Yes / ☐No / ☐NA	
2D and Color Three Chamber	☐Yes / ☐No / ☐NA	
General Comments:		

☐ **Adequate ECHO** ☐ **Inadequate ECHO** ☐ **RESEND NEEDED**

Image Quality Rating: Choose an item.

Data of site notification: Click here to enter a date. **by phone** ☐ **fax** ☐ **e-mail** ☐

Site Contact_____

ECHO-SOL V2 Cardiac Echo Reading Center Review Form

Study Site:			
HCHS-SOL ID: **Reviewer Initials:**			
Date: Choose a date			
	YES	NO	COMMENT
File Format/Upload	☐	☐	
Good EKG Signal	☐	☐	
Acquisitions	☐	☐	
- sinus rhythm (up to 90 bpm): 3 beats	☐	☐	(1-2 beats)
- a-fib or flutter: 5 seconds	☐	☐	
Sweep Speed - 100 mm/ms	☐	☐	(50 mm/s)
Color Doppler - between 50 - 60 cm/sec	☐	☐	
Depth - between 13 - 16 cm	☐	☐	
Parasternal Long Axis			
LV oriented horizontally	☐	☐	
Focus approximately at the level of the IVS	☐	☐	
Decreased depth optimizing LV	☐	☐	
Zoom on the mitral valve	☐	☐	
Zoom on the aortic valve and left ventricular outflow tract	☐	☐	
Color Doppler of aortic and mitral valve	☐	☐	
Parasternal Short Axis			
Short Axis	☐	☐	
- Great vessels --> aortic, pulmonic and tricuspid valves	☐	☐	
- Mitral valve	☐	☐	
- Papillary muscles	☐	☐	
- Apex	☐	☐	
Color Doppler of aortic, tricuspid and pulmonic valves	☐	☐	
Pulsed Doppler of right ventricular outflow tract	☐	☐	

Apical 4			
2D image with endocardial definition	☐	☐	
Decreased depth optimizing LV and LA	☐	☐	
Color Doppler of mitral and tricuspid valves	☐	☐	
CW Doppler of TR	☐	☐	
Pulsed Doppler of MV inflow at leaflet tips	☐	☐	
CW Doppler of the mitral valve inflow	☐	☐	
Tissue Doppler of lateral, septal wall	☐	☐	
Pulsed Doppler of right upper pulmonary vein	☐	☐	
Tissue Doppler of anterior TV leaflet annulus	☐	☐	
M-mode of Tricuspid Annulus	☐	☐	

Apical 2			
2D image with endocardial definition	☐	☐	
Focus approximately at the level of the MV	☐	☐	
Color Doppler of mitral valve	☐	☐	
CW Doppler of MR	☐	☐	
Decreased depth optimizing LV	☐	☐	
Apical 5			
2D image with endocardial definition	☐	☐	
Focus approximately at the level of the MV	☐	☐	
Color Doppler of aortic valve	☐	☐	
CW Doppler of aortic valve	☐	☐	
Pulsed Doppler of left ventricular outflow tract	☐	☐	
Pulsed Doppler of mitral inflow and LVOT - IVRT	☐	☐	
Apical 3			
2D image	☐	☐	
Color Doppler of aortic valve	☐	☐	
Color Doppler of mitral valve	☐	☐	
Comments:			

XI. Instructions for Electronic Transfer of Studies to the Reading Center

Echocardiograms will be transferred from Field Centers to the Reading Center electronically using a secure web-based image transfer technology. A dedicated workstation at each Field Center with high-speed internet capability will be set up for study transfers.
Requirements: Each PC will need a dedicated workspace that is physically secure and also will need a static IP address assigned by each site's IT support. Where available, the PCs will be shipped with the static IP address that the sites provide to WFSM support. The site's firewalls will need to allow for outbound traffic to our electronic **picture archiving and communication systems** (**PACS**).

The program used to provide secure transmission of DICOM images over the internet to the our WFSM PACs is Clinical Trial Processor (CTP), http://mircwiki.rsna.org/index.php?title=CTP-The_RSNA_Clinical_Trial_Processor

CTP will be configured to write the DICOM images from the iE33s to the computer's hard drive. Once CTP recognizes that the images are stored in DICOM format, an anonymizer stage is activated which anyonomizes the data for transport over the internet via an HTTPS protocol. The transfer will take place automatically. Each Field Centers will retain a copy of each echocardiogram, stored on the hard drive of the provided secured pre-configured PC. Due to space constraints, images on the PC will be automatically deleted after 60 days.

Technical Configuration:

a. On the iE33

After each Scan, on the Device Selection Screen, choose WFSM_CTP, this will send the scan via DICOM to our Transmit computer which will automatically store the DICOM image on its hard drive and transmit a copy to our Site.

Press the SETUP key
Go to Print/Network
Go to the Global Configuration tab
Under devices, click NEW DEVICE
PICK ARCHIVE SERVER
Device Name: WFSM_CTP
AE Title: WFSM_CTP
PORT: NEED PORT FOR CTP
IP Address: Static IP address provided by site for the PC

b. For the Sonos 5500

A Crossover cable will be needed to make a connection to the PC to transmit data.

c. For sites without an iE33 device

DICOM image files from the echo machines must hard copied directly onto a CD and be put into a folder on the PC desktop called DICOM, where CTP will look for files and then transmit those to WFSM.

Via Field Center training, sonographers will be instructed on the procedure for submitting a study and tracking documentation directly to the Reading Center electronically. For technical questions regarding study transmission, the Reading Center has an established "ECHO-SOL IT" channel of communication via e-mail.

The process will work as outlined below:

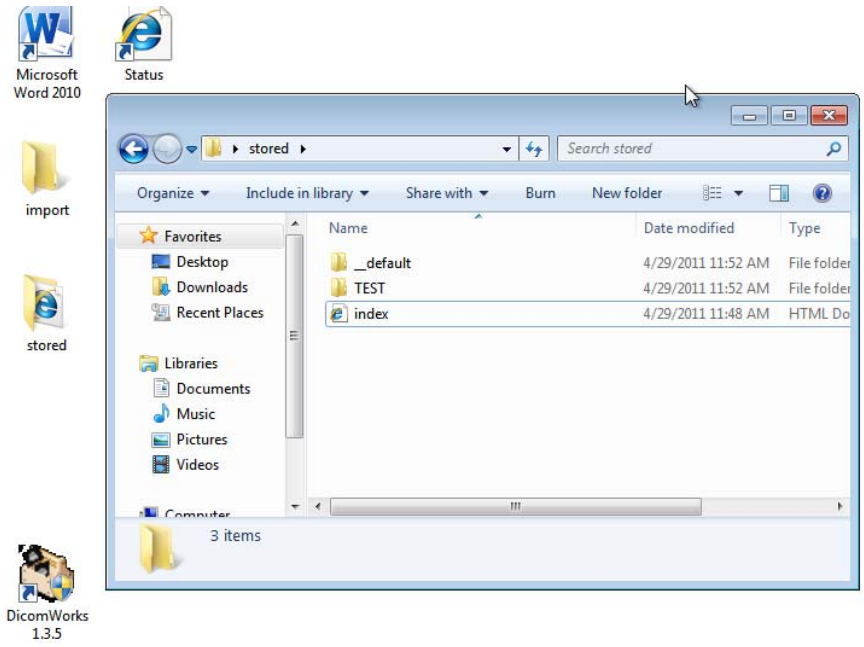
V. Echo Transfer Guide

The iE-33 Echo Machine will need to be configured to send to the CTP PC, for instructions on this, see Technical Configuration.

1. At the Echo Machine, after the study is complete, select the WFSM_CTP archive server from the list of locations to send the study to.

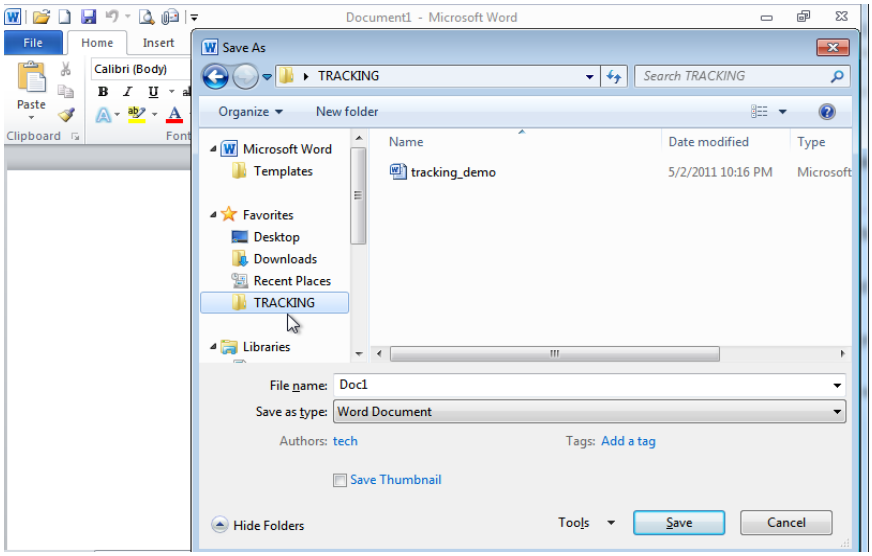
2. After sending the study, the image transfer process is automated. What happens is the PC picks up the images from the iE33, it puts them into a STORED folder (this folder is on your desktop), and it then transmits those images securely over the internet using HTTPS to our server here at WFSM.

If for some reason you have doubts that the study has been sent to the PC, first check the STORED folder



3. Although the digital image data transfer process is automated, after submitting a study, we require that a Data Tracking Form is sent to CERC accompanying every study. You must fill out the Data Tracking Form that is located on the DESKTOP of the CTP PC for every study. This .DOTX MS Word file is created so that when you hit SAVE it will ask you to save the file into a folder. Name the

file using the HCHS-SOL ID, and save it into the TRACKING folder that is on the desktop of the PC.

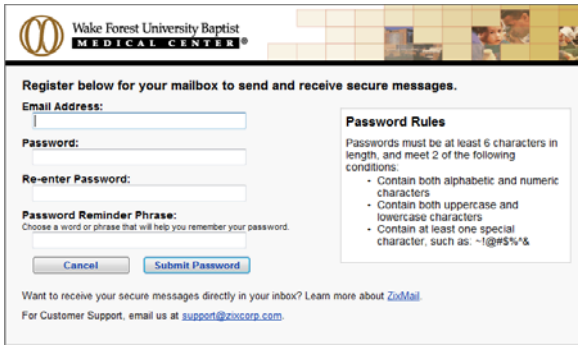


4. To transmit the form to WFSM, you will be using WFSM's SecureMail™ site, this encrypts your mail and send us the form securely. Any replies using this method will be to the email account you specify in the setup.

   a. Go to https://securemail.wfubmc.edu and click Create New Account

   b. Type in your institutional email address (or an email that you check regularly) and create a password of your choosing

   c. After typing the above information, a confirmation email address will be sent to your email account, this account will be displayed as the FROM when you send mail to WFSM.



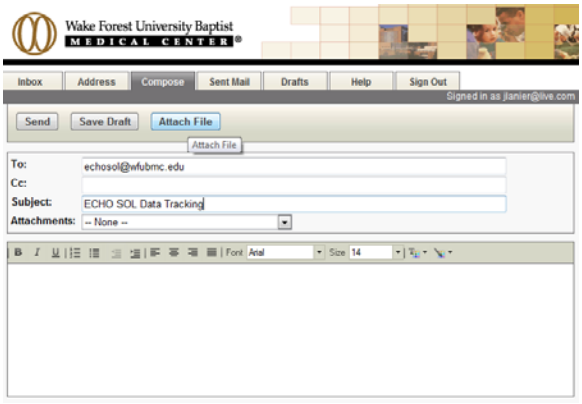
Sending a Data Tracking Form

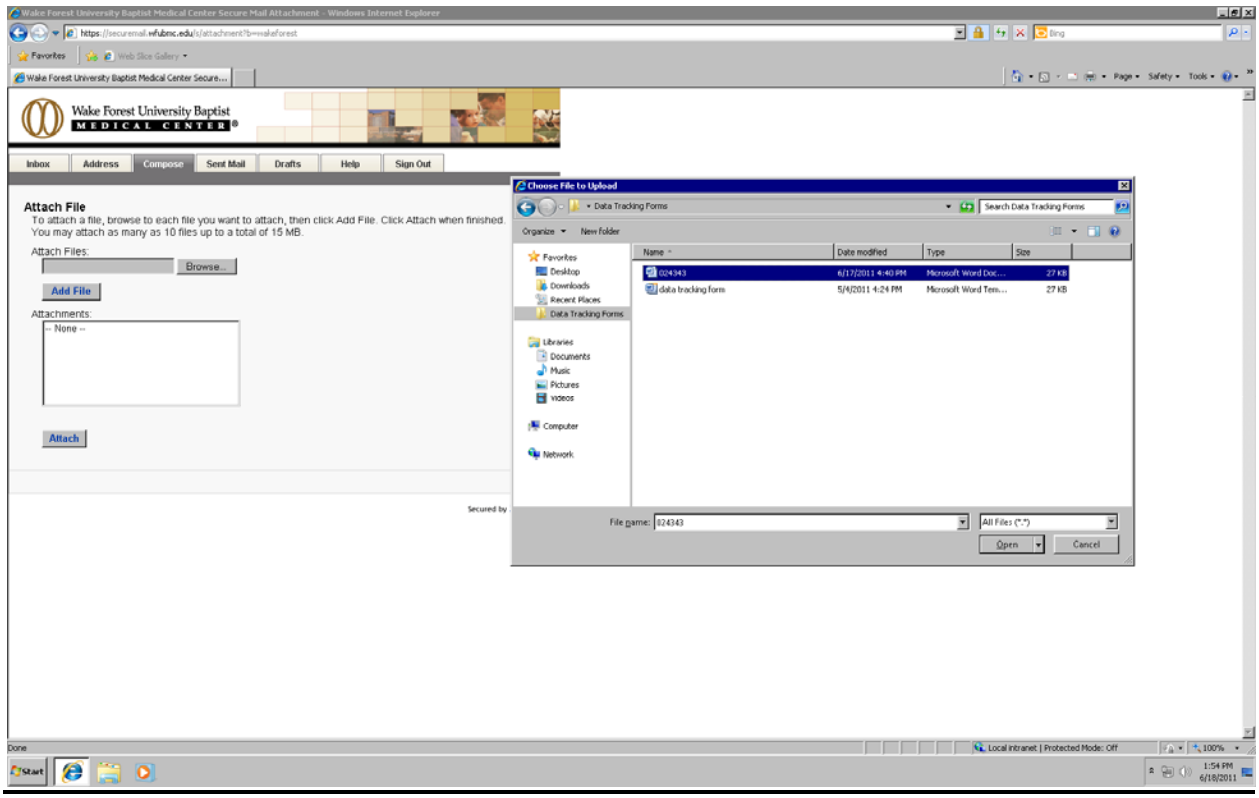
5. Login at https://securemail.wakehealth.edu and click on COMPOSE

6. In the TO field, put echosol@wakehealth.edu

7. For the subject, put "New Transfer"

8. Click on Attach File, then click Browse, to find the TRACKING folder on the desktop, select the tracking form you wish to send and hit OPEN and then ADD FILE





a. If you have another scan you'd like to attach, repeat the above step. When finished, click ATTACH

9. In the body, enter the following:

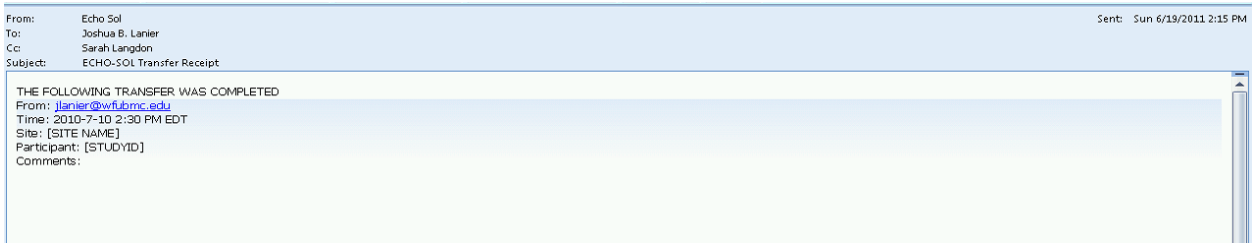
Site: [Site Name]

Participant(s): [STUDY ID]

Comments:

10. Click SEND to send to the Reading Center, you will receive confirmation in your institutional mailbox that the scan has been received.

Transfer email will look like this:



Summary: Instructions for Sending Echos to the Reading Center

1. RECORD ECHO

- Echo should be recorded as per protocol
- No patient identifiers should appear on the echo

2. HIT SEND / WEB TRANSFER

- Send the study from your Philips/HP machine
- The study will transfer to the PC and then automatically transfer via the web to the Core Lab PACS server
- A local copy of the study will remain on the PC for 30 days

3. COMPLETE ECHO TRACKING FORM

- Fill out all fields on the electronic Echo Tracking form and send via e-mail to the Core Lab.