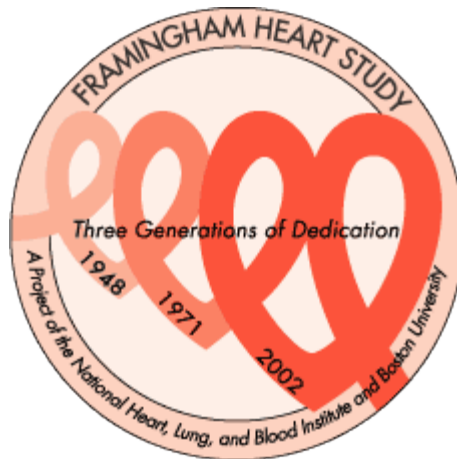


Dataset: t_livrvcte_ex03_3b_1253s

FRAMINGHAM HEART STUDY



Transient Elastography

MANUAL OF PROCEDURES FOR THIRD GENERATION, OMNI 2 AND NEW OFFSPRING SPOUSE EXAMINATION 3

Date: 10/07/2016

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- **Overview**

Participants in the Offspring and Third Generation Cohorts have undergone measurement of liver fat as a part of the multi-detector computed tomography sub-studies. However, the examination of liver fat on CT is insensitive to mild liver fat and also insensitive to liver fibrosis.

All eligible participants at the Third Generation, Omni group 2, and New Offspring Spouse cohorts, 3rd examination will be offered **vibration-controlled transient elastography (VCTE)** of the liver (FibroScan, Echosens Corporation, France). VCTE produces fast, accurate, and noninvasive measurements of liver fat and fibrosis.

- **Background**



www.myliverexam.com/en/lexamen-fibroscan.html

The FibroScan system is an active, non-implantable medical device using ultrasound.

The FibroScan system is intended to provide 50 Hz shear wave speed measurements and estimates of tissue stiffness as well as 3.5 MHz ultrasound coefficient of attenuation parameter (**CAP: Control Attenuation Parameter**) in internal structures of the body.

The FibroScan probe comprises a single-element ultrasound transducer mounted on the shaft of the electrodynamic transducer. This transducer generates a transient vibration, which in turn generates an elastic shear wave. This wave propagates through the skin, the subcutaneous tissues, and then the liver. During shear wave propagation, the ultrasound transducer performs a series of ultrasound acquisitions (emission / reception) to measure the speed of shear wave propagation (V_s) in m/s (**LSM: Liver Stiffness Measurement**). This measurement corresponds to the spatial and temporal average speed of propagation of the shear wave through the liver region of interest, which can be approximated by a cylinder with a diameter of 1 cm and a length of 4 cm (which corresponds to about 3 cm³).

In addition, assuming that the liver is a pure elastic, linear and isotropic medium, the device converts shear wave speed V_s into equivalent stiffness E in kPa using the equation $E = 3 \times \rho \times V_s^2$ with ρ the medium density assumed to be 1000 kg/m³. The values for shear wave speed and equivalent stiffness (or Young's

modulus) are relative indexes intended only for the purpose of comparison with other measurements performed using FibroScan devices.

Concomitantly, the ultrasound acquisitions are used to assess the CAP. Ultrasound attenuation corresponds to the loss of energy as ultrasound propagates through the medium. Due to attenuation, the intensity of emitted ultrasound decreases exponentially with depth (z): $I_z = I_0 \times \exp(-\alpha(f) \times z)$ where I_z is the ultrasound intensity at depth z , α is the frequency (f) dependent attenuation coefficient. Ultrasound attenuation depends principally on (i) the ultrasound frequency, (ii) the properties of the medium of propagation. CAP assesses the value of α at the frequency $f = 3.5$ MHz and is expressed in dB/m.

Absolute values for these measurements may vary among measurement devices produced from different manufacturers.

- **Staff training requirements**

Training requirements for new staff

- 1) Read and understand the manual of procedures.
- 2) Complete training certification program with Echosens representative.
- 3) Perform unprompted VCTE in 5 non-participant volunteers while observed by certified technician or principal investigator.
- 4) Perform VCTE on 5 FHS participants while supervised by certified technician or principal investigator.

- **Subject selection for VCTE**

All participants will be offered VCTE unless they meet any exclusion criteria which will be determined by the technician.

Participants will be excluded if:

- a. They have active medical implants (such as pacemakers, defibrillators, pumps, etc).
- b. They have a wound on their chest wall over the liver which would interfere with probe placement
- c. They are pregnant
- d. They have significant ascites

- **VCTE Equipment and technical support contact**

Fibroscan® 502 Touch with M⁺ and XL⁺ probes

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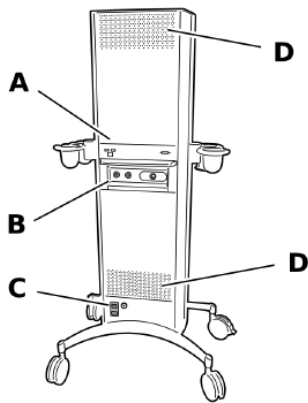
- **VCTE equipment daily procedures**

Please refer to user manual for more detailed instructions and diagrams

- 1) Turn on device (switch labeled “C” below).

5.4. REAR VIEW

The following figure presents the instrument's different user-accessible parts.



Rear view: A: Computer sockets. B: Probe sockets. C: Power connector. D: Perforated ventilation plates.

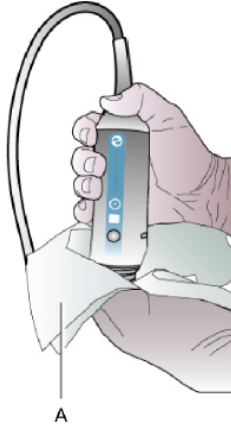
- 2) Clean each probe (see user manual section 8 and instructions below from user manual).



It is not necessary to switch off the device before cleaning the probe.

Surfaces must be cleaned in strict compliance with the following procedures:

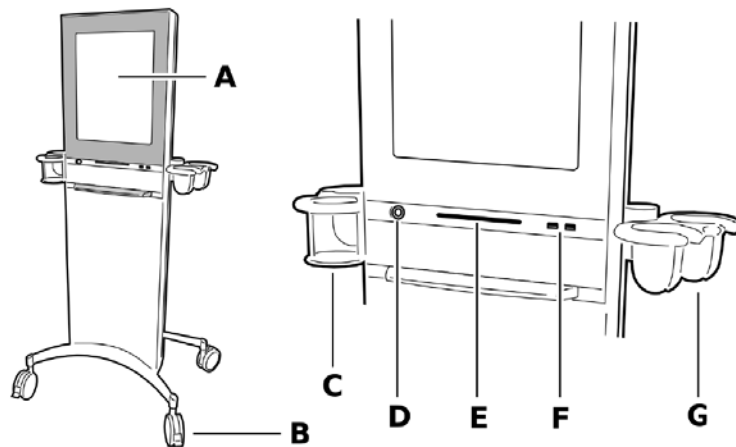
1. Gently remove the gel using a soft cloth or wipe.



Cleaning the probe: A: Wipe.

2. Remove all traces of bodily fluid by cleaning the surfaces using a soft cloth or wipe soaked in the recommended cleaning product.
 3. If necessary, rinse the cleaned surfaces using a soft cloth soaked in water.
 4. Dry, if necessary, using a dry cloth.
 5. Wipe the surfaces using a soft cloth or wipe soaked in the recommended decontamination solution.
 6. Dry, if necessary, using a soft dry cloth.
 7. Examine the transducer and probe cable for any damage such as cracks, breakage, or liquid leakage.
-
- 3) Change file transfer setting to “Data Anonymization – Disabled”
 - a) Choose Fibroscan –Administrator
 - b) Put in admin password: 5UP374DM1N
 - c) Select wrench on the bottom of the screen
 - d) Re-enter admin password: 5UP374DM1N
 - e) Touch the “connectivity icon”
 - f) Choose directory
 - g) Change “Data Anonymization” to Disabled
 - h) Choose Apply
 - i) Exit administrative mode
 - j) NOTE: This must be done any time the machine is turned off or put into standby
 - 4) Date the participant list to serve as the log page for the day.

- 5) Record any/all comments if issues arise or if participant refuses test or has to stop test.
 - a) Record if FibroScan preformed before or after CPET
 - b) Note any food eaten and the time of ingestion
 - c) Note any contraindications
- 6) Between sessions, turn the machine off by pressing the standby button on the bottom left hand corner of the monitor (see label “D” below).



General arrangement of the FibroScan: A: Touch screen. B: Caster with brake. C: Gel holder. D: Standby button with power indicator. E: CD-ROM / DVD-ROM drive. F: Computer sockets. G: Probe holder.

- 7) After testing is complete for the day, save data for each participant onto secure thumb drive (see user manual section 6.7).
- 8) Upload new data to secure server.
- 9) Clean each probe (see user manual section 8).
- 10) Press the standby button to turn off the FibroScan and cut the power supply by setting the main switch to 0.

- **VCTE equipment calibration protocol**

The FibroScan devices will be sequentially calibrated on a yearly basis by Echosens.

When an exam is opened, a window displays the expiry of the calibration of the probe. When this is displayed, the technician or principal investigator will contact Echosens to arrange calibration (service@echosens.com).

- **Participant testing – detailed procedures**

- a. Exclusion criteria

Prior to all tests, the technician must ask the participant about VCTE exclusion criteria:

- Ask: Do you have any implanted medical devices such as a pacemaker? If the device is active, do not perform VCTE.
- Ask: Do you have any wounds or bandages on your chest or abdomen? If yes, determine if the location is over the liver. If any skin defect would interfere with placement of the VCTE probe or patient comfort during the test, do not perform VCTE.
- Ask: Have you been told you have any problems with your liver by your doctor? If the participant answers yes to this question the technician will ask what the underlying liver problem is and make a note of this on their exam. The technician will then ask if the participant has been told that they have excess fluid in their abdomen, called ascites. If yes, the technician will determine if the participant has a large amount of ascites which would interfere with the measurement. If significant ascites is present, the technician will not perform VCTE.
- Ask: Have you had anything to eat or drink today? If yes, note what was eaten and the approximate time it was eaten (or how many hours or minutes previously). Ideally the participant has fasted for at least 2 hours prior to the examination and any snacks will be noted.
- If the participant is a woman of child bearing age, ask if she is pregnant or trying to get pregnant. Pregnant participants will be excluded. While VCTE will not expose the participant to radiation, the device was not originally studied in pregnant women. If the participant reports that she has had tubal ligation or a hysterectomy, has gone through menopause or is infertile, a pregnancy test will not be required, and the participant will be eligible to participate in the VCTE study. All others will be given a pregnancy test. If the pregnancy test is negative, the participant may proceed with the VCTE study. If the test is positive, the participant will be given the result and advised that she may complete the VCTE study after pregnancy. Participants will be asked about possible pregnancy at the time of booking a clinic appointment by FHS personnel. Additional details regarding the pregnancy screening protocol are attached in the addendum.

(To be placed in VCTE testing room)

VCTE EXCLUSION CRITERIA

- DO YOU HAVE ANY IMPLANTED MEDICAL DEVICES SUCH AS A PACEMAKER?
- DO YOU HAVE ANY WOUNDS OR BANDAGES ON YOUR CHEST OR ABDOMEN?
- HAS YOUR DOCTOR TOLD YOU ABOUT ANY PROBLEMS WITH YOUR LIVER?
- HAVE YOU EATEN ANYTHING TODAY?
- ARE YOU PREGNANT OR TRYING TO GET PREGNANT?

b. Description of VCTE test

The technician will describe the test to all eligible participants. A detailed script for the description of the test is included below.

“We are asking you to participate in a study that will take measurements of your liver using a device called “transient elastography or FibroScan”. This device measures the stiffness of the liver and the amount of fat in the liver. Investigators believe liver stiffness and fat may be related to heart disease. If you choose to participate, we will ask that you lie on your back while we use a probe placed on the skin over your liver to take 10 measurements of the liver. The test is painless, but you will feel pressure from the probe during the test. We expect the test to take about 5-10 minutes.”

c. Entering participant information

Note: The FHS reference ID will be entered per participant in the “code” box (see picture below from user manual). The participant name and gender will also be entered. The Tech ID will be entered under “Operator”.

Please see section 6 of the user manual for detailed procedure.

6.5. THE PATIENT RECORD SCREEN

If the patient exists in the patient list, the data will be displayed automatically after the name is entered. Select the patient.

The screenshot shows the 'Patient information' form on the left and a list of patients on the right. The form fields include Lastname, Firstname, Birth date (m/d/yyyy), Gender (F/M), Code, Admitting diagnosis, Operator, Referring physician, and Exam options (CAP, Raw). The patient list on the right shows a grid of patient records with columns for Name, Birth date, and Code. A yellow circle highlights the 'KATZ' patient record.

Name	Birth date	Code
BEZ Kat	12/18/1994	REF4
CHAEI Michael	12/12/1952	REF4
DOE John	3/21/1972	REF2
DUJARDIN Michella	1/12/1982	REF7
KATZ Robin	7/10/2003	REF6
NAPO Léon	3/2/2012	ONIT/UKA
PUBLIC Henry	5/3/1921	REF3
ROE Jane	1/12/1982	REF1
WAN Suzy	6/26/1944	REF5

Complete the fields. At least the 'Name' or 'Code' field must be filled in to start the examination.

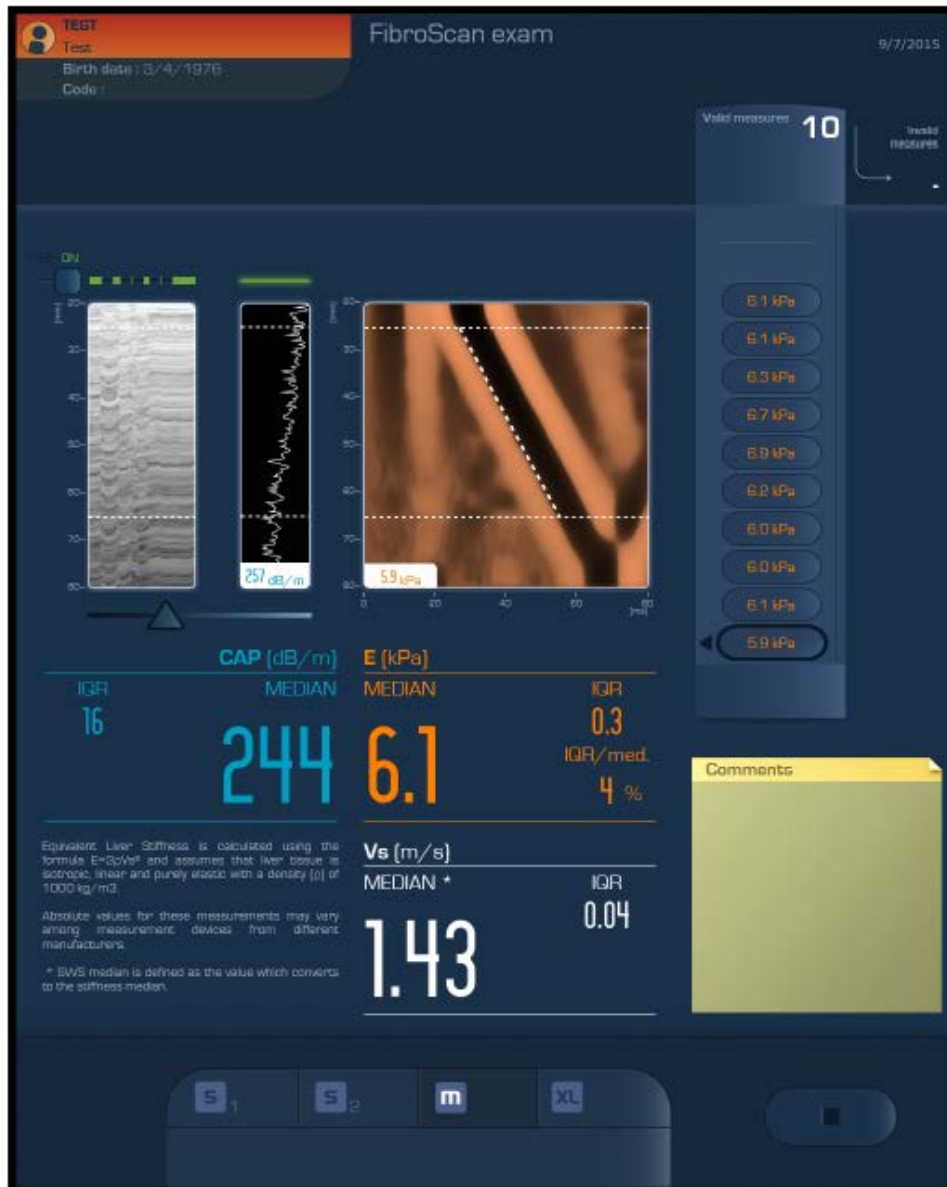
- CAP** Enables/disables the CAP option for the exam about to start.
- Raw** Enables/disables the saving of Raw Data for the exam about to start (available only if Clinical Mode was enabled in the Configuration).

d. Performing VCTE

Please see the user manual for detailed instructions (section 6). A picture of the acquisition screen is displayed below:

6.6. ACQUISITION SCREEN

The main data displayed in an acquisition window are presented below.



The technician will position the participant on the examination table with the FibroScan device on the right side of the participant. Participant will cross right leg over left ankle, and place right arm above head. The M⁺ probe will be used as the default probe for all examinations. A small amount of gel will be placed on the tip of the probe and the probe will be positioned over the liver. Upon contacting the participant's skin, the automatic probe selection tool will begin determining the proper probe for the examination. The results of the automatic probe

selection tool will be displaced on the bottom of the screen and may be one of the following three cases:

1. Recommended probe: in progress: the tool cannot currently measure the SCD because the probe is not correctly positioned in front of the liver and/or the ultrasound signals are of poor quality.
2. 'Recommended probe: The tool measures a SCD that justifies the use of the M+ probe. The box containing the M exam type icon flashes.
3. 'Recommended probe: XL: The tool measures a SCD that justifies the use of the XL+ probe. The box containing the XL exam type icon flashes.

If the tool is able to recommend a probe (case 2 or 3), two situations are possible:

The probe being used matches the recommendation: the operator continues the exam without changing probes and, if he/she wishes, can confirm the probe choice by pressing the icon of the exam type concerned. The probe recommendation tool is then disabled until the end of the current exam.
The probe being used does not match the recommendation: the operator replaces the probe as explained below.

To change the probe during an exam:

1. Select the new probe type. The following message is displayed:
Change of exam type. Changing exam type causes all previous measurements to be definitively erased.
2. Click OK (warning: all measurements taken using the previous probe will be deleted), the following message is displayed: Exam type has changed. Plug the appropriate probe to continue.
3. Plug the appropriate probe if necessary and resume the exam.

After the proper probe is selected, the technician will check the liver targeting tool on the acquisition screen. The color ranges from black (quality of the ultrasound signal is low) to green (quality of the ultrasound signal is high). The greener the color of the liver indicator, the higher the quality, and the better the position of the measurement. Next the technician will check the pressure indicator. When the correct pressure is applied, the indicator will be green. If pressure is too low, the indicator will be orange and if pressure is too much the indicator is red. The measurements cannot be taken if the pressure indicator is not green.

When a measurement is complete, the shear wave propagation map is automatically displayed on the acquisition screen. It represents the levels of liver deformation generated by the propagation of the shear wave as a function of time (horizontal axis in milliseconds) and depth (vertical axis in millimeters). The

shear wave speed value is displayed if the measurement is valid. The color scale indicates the sign of the deformations (compression or expansion). Black areas correspond to negative deformation and pale areas to positive deformation. The black strip through the image represents deformations associated with the passage of the shear wave, which penetrates progressively deeper with time. Invalid measures are automatically rejected by the algorithm if the pulse sent out by the transducer could not be delivered successfully and/or if the shear wave propagation maps are not satisfactory. The message “INVALID” will be displayed above the shear wave propagation map.

Median:

Shear wave speed is expressed in meters per second (m/s), liver stiffness is expressed in kilopascals (kPa) and Control Attenuation Parameter (CAP) is expressed as decibels per meter (dB/m). These values are the median of all valid measurements performed during the examination. CAP is only calculated with the liver stiffness is valid.

If the repeat measurement is invalid, the median is not re-computed. To obtain a reliable and representative liver shear wave speed and liver stiffness measurement, **at least ten valid measurements should be made.**

Interquartile Range (IQR):

The interquartile range (IQR) is expressed in meters per second (m/s), kilopascals (kPa), or decibels (dB/m). It represents the interval around the median within which will fall 50% of all valid measurements. It is recomputed after each new valid measurement.

IQR/Median:

This value, expressed as a percentage, is the ratio of the IQR to the median stiffness. It is re-computed after each new valid measurement. **The goal will be to have the IQR/Median $\leq$ 30% which indicates a high quality examination.**

If the IQR/Median is $> 30\%$ or if a liver stiffness measurement is clearly spurious, the measure can be deleted. The technician will press the measurement to be deleted on the touch screen and press the “X” button to delete. A confirmation request is displayed and indicates the number of measurements if deletion is confirmed. The technician can then continue to take measurements to get to a total of 10.

e. Data procedures after participant testing

- After each examination, the technician will note if any criteria for results reporting are met (see section 10. Results reporting) and will alert the study PI (Michelle Long, MD).

- At the end of the day, all results for that day need to be exported to a removable USB storage device in .XLS format.
- The .XLS file will be transferred to the folder <\\fhs-svr-filserve\Fibroscan> on a daily basis.

Log book: All participants are entered into the “VCTE daily log and comments” binder (including those that have not had a VCTE done so that the reason can be noted). The participant list for the day will serve as the log page. The technician will record any issues with the VCTE as noted below:

ID	VCTE	0=Not Done, 1=Done, 2=Attempted, not finished, 3= Attempted, tech aborted ,4=Other
ID	Reason VCTE not done	1=Medical exclusion, 2=Refused, 3=equipment problems 4=Other

- Quality control

Every three months, the PI (Michelle Long, MD) will observe each technician perform transient elastography on a participant. Constructive feedback will be provided as warranted.

The liver stiffness measurements (LSM) and controlled attenuation parameter (CAP) measurements obtained from participants will be evaluated quarterly. We will review age-, sex-, and body mass index-adjusted averages of the LSM and CAP values during the study interval and compare to the prior interval and expected values. We will stratify by whether or not the FibroScan was performed before or after the cardiopulmonary exercise testing station.

- Results reporting

The FibroScan device measures both liver fibrosis and liver fat. Liver fibrosis may be clinically significant and may warrant additional testing. For this reason, we will report to the participant and their primary care doctor if the liver stiffness measurement (LSM) is ≥ 8.0 KPa which may indicate significant fibrosis of the liver.

Specific procedure:

Technician: The technician will review the results of the LSM obtained from each participant. If the LSM is ≥ 8.0 KPa the technician will call the Principal Investigator (Michelle T. Long, MD) at 617-638-8392 and email at mtlong@bu.edu. After discussion with the principal investigator, the technician will generate the participant letter and letter to the primary care doctor (see below).

Principal Investigator: The principal investigator will review all the results from the participant examinations at least once per month. Any participant with an LSM ≥ 8.0 KPa will receive a letter along with other results from the FHS (as specified by the core exam and other ancillary study results reporting mechanisms). The primary care doctor will be alerted in the form of a letter as well (sent along with other FHS results).

Sample letter to participant:

“Dear FHS participant:

At the Framingham Hearst Study visit you recently participated in, you had a test that measured scarring or “fibrosis” of your liver. The results of this test indicate that you may have increased scarring of your liver. This test is not perfect and I suggest that you follow up with your doctor to discuss a plan for repeating the test (or a similar test) to confirm the results. There are many conditions which can cause liver scarring. If you do have liver scarring, your doctor may suggest additional tests to see what might be causing it.”

Sample letter to PCP:

“Dear Dr. XX

Your patient, XX, underwent transient elastography (FibroScan) of their liver. This test was done for research purposes only. The results of the test indicate that your patient may have significant scarring or fibrosis of their liver. Specifically, the liver stiffness value was XX KPa/IQR. I am alerting you regarding this test result so that you can put this result into the clinical context for

your patient. I would suggest repeating this test (or a similar test evaluating liver fibrosis) in the clinical setting to confirm the finding and determine if the Framingham Heart Study result was a false positive. If your patient does have significant liver fibrosis, you should considering a work up to determine the cause of the liver scarring and/or a referral to a hepatologist. Please do not hesitate to contact me if you have questions at 617-638-8392 or contact Maureen Valentino at 508-935-3417.

- Funding mechanism

This study is supported by the Boston University Department of Medicine, Division of Gastroenterology. Additional support for the study is from Echosens, corporation. The study has Third Party Review from the NHLBI.

- Addendum:

1. Framingham Heart Study Pregnancy Screening Protocol

(To be used in conjunction with script and Pregnancy Determination form for all female participants)

1. Using the IRB approved telephone script, call participant to set up appointment or to remind her of the scheduled exam.
2. Advise the participant that pregnant women or women trying to become pregnant should not have the research test.
3. Complete pregnancy screening using the pregnancy determination form.
4. Notify women of childbearing potential (who are neither pregnant nor trying to become pregnant) that a pregnancy test will be required before the research test and that if the pregnancy test is positive she will not be able to have the research test at that time.
5. Ask the participant to cancel the scheduled appointment if she thinks she may be pregnant.
6. File pregnancy determination form in the participant's chart.
7. On the day of the exam, check responses on pregnancy determination form with participant.
If the research test is part of an ancillary study, make a copy of the pregnancy determination form and give it to the ancillary study coordinator for their records.
8. Advise participants who require pregnancy test that pregnancy tests are not always accurate. (They can produce false negative results.) Therefore, if the participant thinks she may be pregnant or if she is trying to conceive, she should not have the test.
9. Complete the pregnancy test.
10. If the test is positive, give the participant the result and let her know that she cannot complete the test while pregnant.

11. If the test is negative and the participant has no reason to believe she may be pregnant, she may have the test.

Pregnancy Screening Telephone Script

(Use when setting up appointment or at time of reminder call for FHS core exam)

“Hello my name is _____. I am calling from the Framingham Heart Study to schedule your appointment / or to remind you of your appointment for the ____ test/FHS Clinic Exam. Women who are pregnant or trying to become pregnant should not have this test to avoid any potential risk to the fetus (unborn child). Therefore, we need to ask you a few questions to be sure that you are not pregnant. “

(Refer to pregnancy determination form.)

If applicable:

“On the day of the test we will complete a pregnancy test. If the test is positive, you will not be able to have the test at this time.

Pregnancy tests are not always accurate. Therefore, if you think you may be pregnant or if you are trying to conceive, please call to cancel the appointment and/or to let us know, as pregnant women should not have this test.

Thank you very much for participating in the Framingham Heart Study. Please do not hesitate to call us if you have any questions regarding the test, or if you are unsure about any of the details that we have just discussed.”

**The Framingham Heart Study
Pregnancy Determination Form
[Required of all Women]**

FHS I.D. Number:

Last Name:

First Name:

Middle Name:

1. Have you had a hysterectomy [removal of the uterus] or tubal ligation [tubes tied] or gone through menopause? (If the participant is unsure what menopause means or asks questions regarding the definition, check don't know and advise participant that we will be completing a pregnancy test.)

- ☐ Yes → Pregnancy test NOT required
- ☐ Don't know → Pregnancy test REQUIRED
- ☐ No → Pregnancy test REQUIRED

2. Are you pregnant or trying to conceive?

- ☐ Yes → Participant cannot have the test.
- ☐ Don't know → Participant cannot have the test.
- ☐ No → Pregnancy test REQUIRED

3. Pregnancy Test Required? ☐ Yes → Result ☐ Positive ☐ Negative
☐ No

4. Date of pregnancy interview and pregnancy test [if required]: _____
m m d d y y y y

5. ID number of the person completing this form: ☐ ☐ ☐ ☐