Framingham Heart Study

Manual of Procedures

MOP-version 1.0

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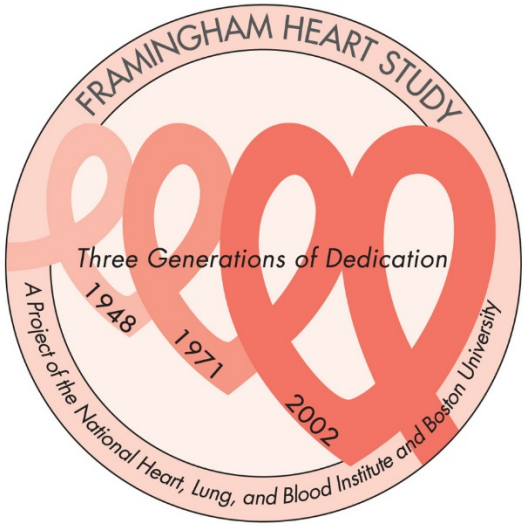
Laboratory: Analytical



Tracking of Revisions to this FHS Protocol MOP

Revised Section	Revision Author	Date (s) of Revisions; source	Approved by, Date	Revisions	Previous Pages #s section changed	Distribution Date

Table of Contents

1. Control and Calibrator Use 4
2. Control Directory 7
3. New Lot Transition 8
4. Roche/Cobas c501 Run 9
5. Roche C501 Weekly Maintenance 12
6. Albumin 13
7. Alanine Transaminase 15
8. Aspartate Transaminase 17
9. Cholesterol 19
10. Serum Creatinine 21
11. C-Reactive Protein 23
12. Glucose 25
13. HbA1c 27
14. Direct HDL 29
15. Triglycerides 31

1.0 Control and Calibrator Use

PRINCIPLE/PURPOSE: To describe the preparation and use of controls and calibrators in the FHS Lab.

1. CONTROL LIST

1.1. Controls are reconstituted according to manufacturers' instructions (see below)

- Controls are labeled with the date of reconstitution or open date
- A volumetric pipette is used for reconstitutions
- Immediately after use, all controls should be returned to storage at 2-8°C

1.2. LYPHOCHEK CHEMISTRY CONTROL LEVELS 1 AND 2 (BIORAD)

- Controls are stored at 2-8°C, allow to reach room temperature (18-25°C) before reconstitution
    - Using a volumetric pipette, reconstitute each vial with 5.0 mL of de-ionized water
    - Replace the stopper and allow the control to stand for 20 minutes, swirling occasionally
    - Transfer contents of 5 vials into 100ml beaker. Allow to mix on stirrer for at least 30 minutes before aliquotting. Approximately 0.4 mL is distributed into separate cryovials. These controls are stored at -80°C and used daily for QC material.
    - The reconstitution date, expiration date and the lot number is recorded on each cryovial, the cryo storage box and in the reagent log book.

1.3. LIQUICHEK LIPIDS CONTROL LEVELS 1 AND 2 (BIORAD)

- Controls are stored at -20°C, need to reach room temperature (18-25°C) before use.
- Do not use warm water or heat to accelerate the process.
- Once the control has reached room temperature, swirl gently to ensure homogeneity.

1.4. LOW CREATININE CONTROL (prepared in house)

- Controls are stored at 2-8°C, allow to reach room temperature (18-25°C) before reconstitution
- Using a 5 ml volumetric pipette and de-ionized water, reconstitute 3 vials of BIORAD LYPHOCHEK® CHEMISTRY CONTROL LEVEL1.
- Mix gently. Let sit for at least 20 minutes.
- Using a transfer pipette, transfer the contents of one of the BR1 vials to a 100 ml volumetric flask.
- Using the same transfer pipette transfer approximately 1-2 ml of de-ionized water to the BR1 vial. Re-cap vial and mix gently. Using the same transfer pipette transfer water in the vial to the volumetric flask. Continue transferring and rinsing vials until all the vials have been rinsed twice.
- Using a transfer pipette, QS the volumetric flask to 100 using de-ionized water.
- Cap the volumetric flask and invert gently to mix.
    - Aliquot 0.4ml of creatinine low control into appropriately labeled cryo vials. These controls are stored at -80°C and used daily for QC material.

- The reconstitution date, expiration date and the lot number is recorded on each cryovial, the cryo storage box and in the reagent log book.
- Depending on the expiration date of the BIORAD Level 1, it may be necessary to lower the total volume of the creatinine low control. Depending on the concentration of the control a different dilution can be used.

1.5. DIABETES LYPHOCHEK CONTROL LEVELS 1 AND 2 (BIORAD)
- Controls are stored at 2-8°C, allow to reach room temperature (18-25°C) before reconstitution
- Reconstitute each vial with 0.5 ml of de-ionized water
- Replace the stopper and allow the control to stand for at least 15 minutes, swirling occasionally
- Before sampling, gently invert the vial several times to ensure homogeneity

1.6. PRECINORM AND PRECIPATH U PLUS (ROCHE)
- Controls are stored at 2-8°C, allow to reach room temperature (18-25°C) before reconstitution
- Open bottle 1, avoiding the loss of lyophilizate, and pipette in exactly 3.0 mL of diluent (bottle 2)
- Close bottle and dissolve the contents completely by occasional gentle swirling for 30 minutes
- Used for supplemental information only.

1.7. PRECINORM L (ROCHE)
- Controls are stored at 2-8°C, allow to reach room temperature (18-25°C) before reconstitution
- Avoiding the loss of lyophilizate pipette in exactly 3.0 mL of de-ionized water
- Close bottle and dissolve the contents completely by occasional gentle swirling for 30 minutes
- Used for supplemental information only

1.8. PRECIPATH HDL/LDL (ROCHE)
- Controls are stored at 2-8°C, allow to reach room temperature (18-25°C) before reconstitution
- Avoiding the loss of lyophilizate pipette in exactly 3.0 mL of de-ionized water
- Close bottle and dissolve the contents completely by occasional gentle swirling for 30 minutes
- Used for supplemental information only

1.9. CRP T CONTROL (ROCHE)
- The controls are ready for use
- Mix carefully before use
- Avoid the formation of foam

2. CALIBRATOR LIST

2.1. CFAS - Calibrator for Automated Systems (ROCHE)
- Controls are stored at 2-8°C, allow to reach room temperature (18-25°C) before reconstitution
- Carefully open bottle, avoiding the loss of lyophilizate, and pipette 3.0 ml of de-ionized water

- Close bottle, dissolve the contents completely by occasional gentle swirling within 30 minutes

2.2 CFAS HbA1c (ROCHE)

- Controls are stored at 2-8°C, allow to reach room temperature (18-25°C) before reconstitution
- Carefully open bottle, avoiding the loss of lyophilizate, and pipette in 2.0 ml of de-ionized water
- Close bottle, dissolve the contents completely by occasional gentle swirling within 30 minutes

2.3. CFAS LIPID (ROCHE)

- Controls are stored at 2-8°C, allow to reach room temperature (18-25°C) before reconstitution
- Carefully open bottle, avoiding the loss of lyophilizate, and pipette in 1.0 ml of de-ionized water
- Close bottle, dissolve the contents completely by occasional gentle swirling within 30 minutes

2.4. CFAS PROTEIN (ROCHE)

- The calibrator is ready for use
- Mix carefully before use
- Avoid the formation of foam

3. RECONSTITUTION AND FREEZING

A few controls and calibrators used in the FHS Lab are prepared and frozen until needed. They are thawed and brought to room temperature prior to use. Once used, the remaining volume is discarded. The controls we freeze are BIORAD LYPHOCHEK CHEMISTRY CONTROL LEVELS 1 AND 2 and LOW CREATININE CONTROL. They are frozen at -80^{o}C. The calibrators we freeze are CFAS, CFAS A1c and CFAS LIPIDS. Calibrators are frozen at -20^{o}C.

(Edition: 05.25.17)

2.0 Control Directory

TEST	MANUFACTURER	CONTROL NAME	ORDER NUMBER
ALBUMIN, ALT, AST,GLUCOSE, HDL	BIO RAD	LYPHOCHECK® ASSAYED CHEMISTRY CONTROL LEVEL 1 LYPHOCHECK® ASSAYED CHEMISTRY CONTROL LEVEL 2	C-310-5 C-315-5
CHOLESTEROL, TRIGLYCERIDE	BIO RAD	LYPHOCHECK® ASSAYED CHEMISTRY CONTROL LEVEL 2 LIQUICHEK® LIPIDS CONTROL LEVEL 2	C-315-5 642
SERUM CREATININE	BIO RAD	LYPHOCHECK® ASSAYED CHEMISTRY CONTROL LEVEL 1 LYPHOCHECK® ASSAYED CHEMISTRY CONTROL LEVEL 2 LYPHOCHECK® ASSAYED CHEMISTRY CONTROL LEVEL1 [1:2.5 Dilution)	C-310-5 C-315-5 C-310-5
CRP	BIO RAD	LIQUICHEK® LIPIDS CONTROL LEVEL 1 LIQUICHEK® LIPIDS CONTROL LEVEL 2	641 642
HbA1c	BIO RAD	DIABETES (LYPHOCHEK) 1 DIABETES (LYPHOCHEK) 2	740

3.0 New Lot Transition

PRINCIPLE/PURPOSE

To adequately identify analytical changes in new lot of reagents and calibrators

1. **Lot Changes**
    - Record “in use date” in reagent inventory book
    - Recalibrate assay
    - Run and evaluate QC results
    - Obtain and run 5 samples performed on the previous lot
    - Obtain and run 5 previously ran CAP or CDC samples
    - Record information on WS.23.New Lot Recheck
    - Determine acceptable delta and bias of samples
    - Lab supervisor or manager will evaluate and make a determination to accept the new lot

(Edition: 05.02.17)

4.0 Roche/Cobas c501 Run

PRINCIPLE/PURPOSE

Run specific assays on controls and participant samples on automated analyzer Roche Cobas c501.

1. SPECIMEN REQUIREMENTS AND COLLECTION METHOD

1.1. Use plasma, serum, and/or whole blood collected by standard technique. See SOP.Sample Collection
1.2. Sample size for the c501 Roche/Cobas Analyzer is approximately 165 µL.
1.3. Freshly collected sample is optimal.
1.4. Prior to analysis the sample should be free of air bubbles.

2. AUTOMATIC START UP:

The system is programmed to start up automatically each morning. The analyzer is put in sleep mode at the end of the previous working session and a wake-up time is set.

2.1. The system starts up automatically at the set time each morning. While the system is performing initialization (takes approximately 12 minutes), the log-on screen is displayed. The system will go into standby after initialization.

2.2. Enter your operator ID and Password to log on or use the following:

- Generic log on - ID: lab Password: fhslab

3. CHECK SYSTEM ALARMS

- If the Alarm global button on the right of the screen is flashing, check the Alarm screen for important system notifications.
  - Tap Alarm (global button) to display the Alarm screen.
  - Select each alarm to view the description and remedies (displayed on lower half of the screen).
  - Correct any alarm conditions by following the remedies.

4. SYSTEM OVERVIEW SCREEN

The workflow guide area at the top of the system overview screen guides you through the preroutine operation. A color scheme is used to display the status of the system. Green – Standby, Light Blue – (operation or maintenance) Yellow – Caution, Red – an alarm that requires action on the part of the operator.

5. PREROUTINE OPERATION

5.1. Perform requested maintenance. **See SOP Daily Maintenance or SOP Weekly Maintenance**.

5.2. Reagent Preparation

- Return to System Overview Screen (F12) and choose the Reagent Preparing Button.
- Choose Reagent Load/ Unload List on the Reagent Preparing Window. A confirmation window appears.
- Choose yes to print the Reagent Load/ Unload List.
- Replace any required reagents, diluents, detergent or wash solution according to the list. Ensure that all reagents have not exceeded their expiration dates.

6. CALIBRATION AND QC

- It is necessary to calibrate all applications and measure quality control (QC) samples regularly to verify the stability of reagents and the entire system. Make sure calibration has been completed successfully and all QC results are within acceptable limits before you start routine operation.
- Check the Lab Daily QC book and Lab Notes for any recommended calibrations.
- Touch Calibration Key. Manually select calibrations and save. Select highest calibration that is allowed.
- Touch Calibration and QC Select key. Touch Routine QC key. All QC will be activated.

6.1. Calibration

- Place water and appropriate calibrator in assigned rack/position.
- Place rack on rack loader.
- Hit Global Start and then the next Start button that appears.
- Calibration will print out when done.

6.2. QC

- Place controls in assigned racks/positions.
- Hit Global Start and then the next Start button that appears.
- QC will print out when complete. Verify that it is within acceptable ranges.

7. PROGRAM : Bar- Coded Samples

7.1. Touch Workplace. Touch Test Selection
7.2. Choose Sample type (serum/plasma or supernatant for A1C). Enter Sample ID - Enter
7.3. Touch a panel key or individual test key. Save. Repeat if necessary.
7.4. Load barcoded samples into the racks so that the barcode is visible. Position **does not** matter.
7.5. Load racks onto loader. Press Global Start and Start again

8. PROGRAM : NON Bar- Coded Samples

8.1. Touch Workplace. Touch Test Selection
8.2. Choose Sample type (serum/plasma or supernatant for A1C). Enter Sample ID - Enter
8.3. Touch a panel key or individual test key.

8.4. Touch bar code error button. This brings you to a screen where you can assign a rack and position number. Save. Then save again. Repeat if necessary
8.5. Load non barcoded samples into the racks. **Samples must be place in the rack/position as ordered**.
8.6. Load racks onto loader. Press Global Start and Start again

9. RESULTS

9.1. As results are generated on the instrument, they are temporarily saved in the data base located on the internal hard disk of the control unit.
9.2. When all results for all the tests requested for a sample are available, a hard copy is printed with the run results for the days' samples. Print by sample ID.
9.3. To view results: Workplace > Data Review Screen; test results are displayed here.

10. BACKUP DISCS

10.1. Data is back up daily into both a Binary and Ascii File.
10.2. After completion of run, go to System Overview > Sample Data Clear > Back up Clear > Mass Storage > Binary or ASCII (have to check off with double quotation marks and Separation "I") > Ok.
10.3. Binary data is saved on a disk. ASCII data is saved on a removable drive.
10.4. Take ASCII data and upload it to lab 7 > datasets > 1 Daily Cobas download > File year Daily cobas download > Month File > Week File.

11. REFERENCES

See Roche/Cobas Operating Manual for detailed operating instructions.

(Edition 06.06.17)

5.0 Roche C501 Weekly Maintenance

PRINCIPLE/PURPOSE: To ensure the cobas analyzer is functioning properly.
The instrument is programmed to perform the weekly pipe every Monday morning at start up. A maintenance pipe is a set of system-controlled maintenance items. Using maintenance pipes saves time by allowing the system to proceed from one maintenance item to another automatically without operator intervention.

1. **Weekly Pipe:**
   1.1. Daily pipe
   1.2. Wash reaction parts
   1.3. Cell blank measurement

   To view cell blank results hit global Print button—Utility—Cell Blank—Preview—View. Check abnormal cell list for issues. Check off on Maintenance Log.

2. **WEEKLY MAINTENANCE:**
   2.1. Record Checklist Activities on the Roche Cobas 501 Analyzer Maintenance Log
   - All cell covers are to be cleaned with alcohol followed by deionized water. Gently wipe down the lid on both sides with an alcohol pad.
   - Rinse stations are first rinsed with 2% Eco-D solution followed by 100 ml of deionized water. Fill a 100ml syringe to 50ml with the deionized water and inject the solution into the drain hole of the sample probe, reagent probe and ISE probe rinse bath. Make sure the sides of the drain are rinsed as well. Repeat with another 50ml of deionized water. Carefully avoid the drying port at the top of each drain well. Use an alcohol pad to gently dry the top of the dryer port.

3. **REFERENCES**

See Roche/Cobas Operating Manual Sections c-29, c-78 through c-81 for detailed instructions on Weekly Operating Check.

(Edition: 06.06.17)

6.0 Albumin

PRINCIPLE/PURPOSE

For the quantitative determination of Albumin in serum on automated analyzer Roche Cobas c501.

1. SPECIMEN REQUIREMENTS AND COLLECTION METHOD

1.1. Use serum collected by standard venipuncture technique. See SOP.Sample collection

1.2. Sample size for serum panel on the Roche Cobas Analyzer c501 is approximately 165 µL.

1.3. Freshly collected sample is optimal. Prior to analysis the sample should be free of air bubbles.

2. MATERIALS

MATERIAL	ORDER NUMBER	MANUFACTURER
Albumin 2 Reagent	03183688122	Roche
Lyphochek® Assayed chemistry Control Level 1	C-310-5	BioRad
Lyphochek® Assayed chemistry Control Level 2	C-315-5	BioRad

3. CALIBRATION

3.1. Calibrator

CFAS- Calibrator for Automated Systems

3.2. Calibration Frequency

See SOP.Assay Parameter for Albumin Assay.

4. QUALITY CONTROL

Two levels of commercially available control materials with assigned values are used (see 15.1 in Quality Manual). For corrective action of controls outside reference ranges see 15.3 in Quality Manual.

5. AUTOMATED ANALYZER OPERATION – See SOP.Cobas c501 Run.

5.1. ASSAY SELECTION

Select the Albumin Assay by touching the ALB2 key on the screen.

5.2. MAINTENANCE

Maintenance is performed as specified by manufacturer's operation protocols.

6. TEST REPORTING

6.1. Analytical Measurement Range

- Reportable Range: 0.2 – 6.0 g/dL

6.2. Repeat Values

- Repeat Levels: <3.0 g/dL or 6.0 g/dL
- Physician Notification Levels: <3.0 g/dL

6.3. Expected Values

- Adults: 3.5-5.2 g/dL

7. LIMITATIONS OF THE METHOD

See Current Roche Cobas Application Sheets.

8. REFERENCES

See Current Roche Cobas Application Sheets.

(Edition 11.07.16)

7.0 Alanine Transaminase

PRINCIPLE/PURPOSE

For the quantitative determination of ALT in serum on automated analyzer Roche Cobas c501.

1. SPECIMEN REQUIREMENTS AND COLLECTION METHOD

1.1. Use serum collected by standard venipuncture technique. See SOP.Sample collection.

1.2. Sample size for serum panel on the Roche Cobas Analyzer c501 is approximately 165 µL.

1.3. Freshly collected sample is optimal. Prior to analysis the sample should be free of air bubbles.

2. MATERIALS

MATERIAL	ORDER NUMBER	MANUFACTURER
Alanine Transaminase Reagent	20764957322	Roche
Lyphochek® Assayed chemistry Control Level 1	C-310-5	BioRad
Lyphochek® Assayed chemistry Control Level 2	C-315-5	BioRad

3. CALIBRATION

3.1. Calibrator

CFAS- Calibrator for Automated Systems

3.2. Calibration Frequency

See SOP.Assay Parameter for ALT Assay.

4. QUALITY CONTROL

Two levels of commercially available control materials with assigned values are used (see 15.1 in Quality Manual). For corrective action of controls outside reference ranges see 15.3 Quality Manual.

5. AUTOMATED ANALYZER OPERATION – See SOP.Cobas c501 Run.

5.1. ASSAY SELECTION

Select the ALT Assay by touching the ALTL key on the screen.

5.2. MAINTENANCE

Maintenance is performed as specified by manufacturer's operation protocols.

6. TEST REPORTING

6.1. Analytical Measurement Range

- Reportable Range: 5 – 653 U/L

6.2. Repeat Values

- Repeat Level: >50 U/L
- Physician Notification Level: > Males: >90 U/L

  >Females: >57 U/L

6.3. Expected Values

- Males: ≤ 41 U/L
- Females: ≤ 33 U/L

7. LIMITATIONS OF THE METHOD

See Current Roche Cobas Application Sheets.

8. REFERENCES

See Current Roche Cobas Application Sheets.

(Edition 11.01.17)

8.0 Aspartate Transaminase

PRINCIPLE/PURPOSE

For the quantitative determination of AST in serum on automated analyzer Roche Cobas c501.

1. SPECIMEN REQUIREMENTS AND COLLECTION METHOD

1.1. Use serum collected by standard venipuncture technique. See SOP.Sample Collection.

1.2. Sample size for serum panel on the Roche Cobas Analyzer c501 is approximately 165 µL.

1.3. Freshly collected sample is optimal. Prior to analysis the sample should be free of air bubbles.

2. MATERIALS

MATERIAL	ORDER NUMBER	MANUFACTURER
Aspartate Transaminase Reagent	20764949322	Roche
Lyphochek® Assayed chemistry Control Level 1	C-310-5	BioRad
Lyphochek® Assayed chemistry Control Level 2	C-315-5	BioRad

3. CALIBRATION

3.1. Calibrator

CFAS- Calibrator for Automated Systems

3.2. Calibration Frequency

See SOP.Assay Parameter for AST Assay.

4. QUALITY CONTROL

Two levels of commercially available control materials with assigned values are used (see 15.1 in Quality Manual). For corrective action of controls outside reference ranges see 15.3 in Quality Manual.

5. AUTOMATED ANALYZER OPERATION – See SOP.Cobas c501 Run.

5.1. ASSAY SELECTION

Select the AST Assay by touching the SASTL key on the screen.

5.2. MAINTENANCE

Maintenance is performed as specified by manufacturer's operation protocols.

6. TEST REPORTING

6.1. Analytical Measurement Range

- Reportable Range: 5 – 684 U/L

6.2. Repeat Values

- Repeat Level: >50 U/L
- Physician Notification Level: > Males: >90 U/L
  >Females: >57 U/L

6.3. Expected Values

- Males: up to 40 U/L
- Females: up to 32 U/L

7. LIMITATIONS OF THE METHOD

See Current Roche Cobas Application Sheets.

8. REFERENCES

See Current Roche Cobas Application Sheets.

(Edition 11.01.17)

9.0 Cholesterol

PRINCIPLE/PURPOSE

For the quantitative determination of Cholesterol in plasma on automated analyzer Roche Cobas c501.

1. SPECIMEN REQUIREMENTS AND COLLECTION METHOD

1.1. Use EDTA plasma collected by standard venipuncture technique. See SOP.Sample Collection.

1.2. Sample size for plasma panel on Roche Cobas Analyzer c501 is approximately 165 µL.

1.3. Freshly collected sample is optimal. Prior to analysis the sample should be free of air bubbles.

2. MATERIALS

MATERIAL	ORDER NUMBER	MANUFACTURER/VENDOR
R1 Cholesterol Reagent	03039773190	Roche
Lyphochek Assayed Chemistry Control Level 2	C-315-5	BioRad
Liquichek Lipids Control Level 2	642	BioRad

3. CALIBRATION

3.1. Calibrator

CFAS- Calibrator for Automated Systems

3.2. Calibration Frequency

See SOP.Assay Parameter for Cholesterol Assay.

4. QUALITY CONTROL

Two levels of commercially available control materials with assigned values are used (see 15.1 in Quality Manual). For corrective action of controls outside reference ranges (see 15.3 in Quality Manual).

5. AUTOMATED ANALYZER OPERATION – See SOP.Cobas c501 Run.

5.1. Assay Selection

Select the Cholesterol Assay by touching the CHOL2 key on the screen.

5.2. Maintenance

Maintenance is performed as specified by manufacturer's operation protocols.

6. TEST REPORTING

6.1. Analytical Measurement Range

- Reportable Range: 4-769 mg/dL

6.2. Repeat Values

- Repeat Level: <120 mg/dL and >350 mg/dL
- Physician Notification Level: >350 mg/dL

6.3. Expected Values

- Desirable Blood Cholesterol: <200 mg/dL
- Borderline-High Blood Cholesterol: 200 – 239 mg/dL
- High Blood Cholesterol: ≥ 240 mg/dL

6.4. An average of duplicate results are used to obtain value

7. LIMITATIONS OF THE METHOD

See Current Roche Cobas Application Sheets

8. REFERENCES

See Current Roche Cobas Application Sheets

(Edition 07.06.17)

10.0 Serum Creatinine

PRINCIPLE/PURPOSE

For the quantitative determination of creatinine in serum on automated analyzer Roche Cobas c501.

1. SPECIMEN REQUIREMENTS AND COLLECTION METHOD

1.1. Use serum by standard venipuncture technique. See SOP.Sample Collection.

1.2. Sample size for the Roche Cobas Analyzer c501 is approximately 165 µL.

1.3. Freshly collected sample is optimal. Prior to analysis the sample should be free of air bubbles.

2. MATERIALS

MATERIAL	ORDER NUMBER	MANUFACTURER
CREAJ GEN2 Reagent	04810716190	Roche
LYPHOCHECK® ASSAYED CHEMISTRY CONTROL LEVEL1	C-310-5	BIO RAD
LYPHOCHECK® ASSAYED CHEMISTRY CONTROL LEVEL2	C-315-5	BIO RAD
LYPHOCHECK® ASSAYED CHEMISTRY CONTROL LEVEL1 [1:2]	-	BIO RAD

3. CALIBRATION

3.1. Calibrator

CFAS- Calibrator for Automated Systems

3.2. Calibration Frequency

See SOP.Assay Parameter for Serum Creatinine Assay.

4. QUALITY CONTROL

4.1. Two levels of commercially available control materials with assigned values are used (see 15.1 in Quality Manual).

4.2. A low control made from a 1:2 dilution of Level 1 control (see SOP.Control and Calibration Use)

4.3. For corrective action of controls outside reference ranges (see 15.3 in Quality Manual)

5. AUTOMATED ANALYZER OPERATION – See SOP.Cobas c501 Run

5.1. ASSAY SELECTION

Select the Serum Creatinine Assay by touching the CREJ2 button on the screen.

5.2. MAINTENANCE

Maintenance is performed as specified by manufacturer's operation protocols.

6. TEST REPORTING

6.1. Analytical Measurement Range

- Reportable Range: 0.17 – 24.90 mg/dL

6.2. Repeat Values

- Repeat Level: <0.5 mg/dL and >2.0 mg/dL
- Physician Notification Level: >2.0 mg/dL

EXCEPT FOR THOSE PARTICIPANTS THAT ARE UNDERGOING DIALYSIS FOR RENAL FAILURE

6.3. Expected Values

- Adult Males: 0.7 – 1.2 mg/dL
- Adult Females: 0.5 – 0.9 mg/dL

6.4. An average of duplicate results are used to obtain value

7. LIMITATIONS OF THE METHOD

See Current Roche Cobas Application Sheets.

8. REFERENCES

See Current Roche Cobas Application Sheets.

(Edition 07.06.17)

11.0 C-Reactive Protein

PRINCIPLE/PURPOSE

For the quantitative determination of C-Reactive Protein in serum on automated analyzer Roche Cobas c501.

1. SPECIMEN REQUIREMENTS AND COLLECTION METHOD

1.1. Use serum collected by standard venipuncture technique. See SOP.Sample Collection

1.2. Sample size for serum panel on the Roche Cobas Analyzer c501 is approximately 165 µL.

1.3. Freshly collected sample is optimal. Prior to analysis the sample should be free of air bubbles.

2. MATERIALS

MATERIAL	ORDER NUMBER	MANUFACTURER
CRP LX High Sensitive Reagent	04628918190	Roche
Liquichek® Lipids Control Level 1	641	BioRad
Liquichek® Lipids Control Level 2	642	BioRad

3. CALIBRATION

3.1. Calibrator

CFAS- Protein

3.2. Calibration Frequency

See SOP.Assay Parameter for CRP Assay.

4. QUALITY CONTROL

Two levels of commercially available control materials with assigned values are used (see 15.1 in Quality Manual). For corrective action of controls outside reference ranges see 15.3 in Quality Manual.

5. AUTOMATED ANALYZER OPERATION – See SOP.Cobas c501 Run

5.1. ASSAY SELECTION

Select the CRP Assay by touching the CRPHS key on the screen.

5.2. MAINTENANCE

Maintenance is performed as specified by manufacturer's operation protocols.

6. TEST REPORTING

6.1. Analytical Measurement Range

- Reportable Range: 0.15 – 17.72 mg/L

6.2. Repeat Values

- Repeat Level: <0.15 mg/L or >40 mg/L
- Physician Notification Level: N/A

6.3. Expected Values

- Adults: <1.0 mg/L – low risk
- 1.0– 3.0 mg/L – average risk
- >3.0 mg/L – high risk

7. LIMITATIONS OF THE METHOD

See Current Roche Cobas Application Sheets

8. REFERENCES

See Current Roche Cobas Application Sheets

(Edition: 11.07.16)

12.0 Glucose

PRINCIPLE/PURPOSE

For the quantitative determination of Glucose in plasma on automated analyzer Roche Cobas c501.

1. SPECIMEN REQUIREMENTS AND COLLECTION METHOD

1.1. Use EDTA plasma collected by standard venipuncture technique. See SOP.Sample Collection.

1.2. Sample size for EDTA panel on Roche Cobas Analyzer c501 is approximately 165 µL.

1.3. Freshly collected sample is optimal. Prior to analysis the sample should be free of any air bubbles.

2. MATERIALS

MATERIAL	ORDER NUMBER	MANUFACTURER
Glucose HK Gen 3 Reagent	04404483190	Roche
Lyphochek® Assayed chemistry Control Level 1	C-310-5	BioRad
Lyphochek® Assayed chemistry Control Level 2	C-315-5	BioRad

3. CALIBRATION

3.1. Calibrator

CFAS- Calibrator for Automated Systems

3.2. Calibration Frequency

See SOP.Assay Parameter for Glucose Assay.

4. QUALITY CONTROL

Two levels of commercially available control materials with assigned values are used (see 15.1 in Quality Manual). For corrective action of controls outside reference ranges see 15.3 in Quality Manual.

5. AUTOMATED ANALYZER OPERATION – See SOP.Cobas c501 Run.

5.1. ASSAY SELECTION

Select the Glucose Assay by touching the GLUC3 key on the screen.

5.2. MAINTENANCE

Maintenance is performed as specified by manufacturer's operation protocols.

6. TEST REPORTING

6.1. Analytical Measurement Range

- Reportable Range: 2 – 716 mg/dL

6.2. Repeat Values

- Repeat Levels: <70 mg/dL or >125 mg/dL
- Physician Notification Levels: >200 mg/dL (all)
  >300 mg/dL fasting diabetics (alert)

6.3. Expected Values

- <50 mg/dL Hypoglycemia (low blood sugar)
- 50-99 mg/ dL Normal
- 100-125 mg/ dL Borderline Hyperglycemia (borderline high blood sugar)
- ≥126 mg/ dL Hyperglycemia (high blood sugar)

6.4. An average of duplicate results are used to obtain value

7. LIMITATIONS OF THE METHOD

See Current Roche Cobas Application Sheets.

8. REFERENCES

See Current Roche Cobas Application Sheets.

(Edition 07.06.17)

13.0 HbA1c

PRINCIPLE/PURPOSE

For the quantitative determination Hemoglobin A1c in whole blood on automated analyzer Roche Cobas c501. HbA1c determinations are utilized in the long-term monitoring of glycemia.

1. SPECIMEN COLLECTION METHOD

1.1. Use whole blood collected in EDTA plasma tube by standard venipuncture technique. See SOP.Sample Collection

1.2. Freshly collected sample is optimal.

2. SPECIMEN PREPARATION

3.1 Place one EDTA vacutainer per participant on a mixing rocker for 5 minutes.

3.2 Label one 5ml tube for each participant, using a barcoded label.

3.3 Remove the sample from the rocker, avoiding splashing, remove stopper. Transfer 1 ml of well mixed whole blood into the 5 ml tube.

3.4 Recap the vacutainer and process it along with the other EDTA and citrate vacutainers accordingly.

3.5 Cap the 5 ml tube, placing it on the mixing rocker for at least five minutes.

3. MATERIALS

MATERIAL	ORDER NUMBER	MANUFACTURER
HBA1C Gen 3 Tina-quant Reagent	05336163190	Roche
Hemolyzing Reagent	0452818219	Roche
Diabetes Lyphochek® Level 1 and 2	740	Biorad

5. CALIBRATION

3.1. Calibrator

CFAS – HbA1c

3.2. Calibration Frequency

See SOP.Assay Parameter for HbA1c Assay.

4. QUALITY CONTROL

Two levels of commercially available control materials with assigned values are used (see 15.1 in Quality Manual). For corrective action of controls outside reference ranges (see 15.3 in Quality Manual)

5. AUTOMATED ANALYZER OPERATION – See SOP.Cobas c501 Run

5.1. ASSAY SELECTION

» NOTE: BE SURE THAT THE RUN TYPE SELECTED IS SUPRNT!!!!

Select the HbA1c Assay by touching the A1C % key on the screen.

5.2. MAINTENANCE

Maintenance is performed as specified by manufacturer's operation protocols.

6. TEST REPORTING

6.1. Analytical Measurement Range

- %HbA1c : 3.4 – 17.2%
- Hb: 4-40 g/dL
- A1c: 0.3-2.6 g/dL

6.2. Repeat Values

- Repeat Levels: Hemaglobin <6 and >30 g/dL

  HbA1c > current highest calibrator

  HbA1c% ≥6.5% non-diabetic and > 8.0% diabetic
- Physician Notification Levels: ≥6.5% non-diabetic

6.3. Expected Values

- Adults : 4.8 – 5.9%

7. LIMITATIONS OF THE METHOD

See Current Roche Cobas Application Sheets.

8. REFERENCES

See Current Roche Cobas Application Sheets.

(Edition 11.02.17)

14.0 Direct HDL

PRINCIPLE/PURPOSE

For the quantitative determination of Direct HDL in plasma on automated analyzer Roche Cobas c501.

1. SPECIMEN REQUIREMENTS AND COLLECTION METHOD

2.1. Use plasma collected by standard venipuncture technique. See SOP.Sample Collection

2.2. Sample size for plasma panel onthe Roche Cobas Analyzer c501 is approximately 165 µL.

2.3. Freshly collected sample is optimal. Prior to analysis the sample should be free of air bubbles.

2. MATERIALS

MATERIAL	ORDER NUMBER	MANUFACTURER
HDL-C Reagent	04628918190	Roche
Lyphochek® Assayed chemistry Control Level 1	C-310-5	BioRad
Lyphochek® Assayed chemistry Control Level 2	C-315-5	BioRad

3. CALIBRATION

4.1. **Calibrator**

CFAS- Lipid

4.2. **Calibration Frequency**

See SOP.Assay Parameter for Direct HDL Assay.

4. Quality Control

Two levels of commercially available control materials with assigned values are used (see 15.1 in Quality Manual). For corrective action of controls outside reference ranges see 15.3 in Quality Manual.

5. AUTOMATED ANALYZER OPERATION – See SOP.Cobas c501 Run.

5.1. **ASSAY SELECTION**

Select the Direct HDL Assay by touching the HDLC3 key on the screen.

5.2. **MAINTENANCE**

Maintenance is performed as specified by manufacturer's operation protocols.

6. TEST REPORTING

6.1. **Analytical Measurement Range**

- Reportable Range: 3-121 mg/dL

7. Repeat Values

- Repeat Levels: >90 mg/dL
- Physician Notification Levels: <20 mg/dL

8. Expected Values

8.1. Undesirable HDL: <40 mg/dL

9. LIMITATIONS OF THE METHOD

See Current Roche Cobas Application Sheets

10. REFERENCES

See Current Roche Cobas Application Sheets.

(Edition 07.06.17)

15.0 Triglycerides

PRINCIPLE/PURPOSE

For the quantitative determination of Direct HDL in plasma on automated analyzer Roche Cobas c501.

1. SPECIMEN REQUIREMENTS AND COLLECTION METHOD

2.4. Use plasma collected by standard venipuncture technique. See SOP.Sample Collection

2.5. Sample size for plasma panel onthe Roche Cobas Analyzer c501 is approximately 165 µL.

2.6. Freshly collected sample is optimal. Prior to analysis the sample should be free of air bubbles.

2. MATERIALS

MATERIAL	ORDER NUMBER	MANUFACTURER
HDL-C Reagent	04628918190	Roche
Lyphochek® Assayed chemistry Control Level 1	C-310-5	BioRad
Lyphochek® Assayed chemistry Control Level 2	C-315-5	BioRad

3. CALIBRATION

4.3. **Calibrator**

CFAS- Lipid

4.4. **Calibration Frequency**

See SOP.Assay Parameter for Direct HDL Assay.

4. Quality Control

Two levels of commercially available control materials with assigned values are used (see 15.1 in Quality Manual). For corrective action of controls outside reference ranges see 15.3 in Quality Manual.

5. AUTOMATED ANALYZER OPERATION – See SOP.Cobas c501 Run.

5.2. **ASSAY SELECTION**

Select the Direct HDL Assay by touching the HDLC3 key on the screen.

5.2. **MAINTENANCE**

Maintenance is performed as specified by manufacturer's operation protocols.

6. TEST REPORTING

6.2. **Analytical Measurement Range**

- Reportable Range: 3-121 mg/dL

7. Repeat Values

- Repeat Levels: >90 mg/dL
- Physician Notification Levels: <20 mg/dL

8. Expected Values

8.1. Undesirable HDL: <40 mg/dL

9. LIMITATIONS OF THE METHOD

See Current Roche Cobas Application Sheets

10. REFERENCES

See Current Roche Cobas Application Sheets.

(Edition: 07.06.17)