

Testing Samples with FaSafe™ HSY Kit

Step 1: Preparing Reagents

- Remove FaSafe™ Reagents A, B, calibrators, controls and Preparation Reagent from the refrigerator and allow them to reach room temperature (18–25°C). This should take approximately 2 hours.
- Power ON the SafTest Analyzer by pressing the [I] marked portion of the power switch, which is located on the rear panel of the SafTestII Analyzer. Allow 5 minutes to warm up.
- Preheat the heat block to 37–42°C. Use temperature setting LOW and dial #4. This is an approximate setting, check with thermometer to verify. Adjust dial (0-10) to correct for the right temperature.



Step 2: Setting up the Calibration Curve

When you receive a new kit you must first run a calibration curve

- Label 5 new 12mm glass test tubes to correspond to the Calibrator 1, 2, 3 and 4; and 2 new 12mm tubes for duplicates of the Control.
- Mix amber bottles containing the calibrators and controls to ensure you have a homogenous solution.
- Use a positive displacement pipette to transfer 100µl of the calibrators into the corresponding labeled test tube.
- Use a positive displacement pipette to transfer 100µl of the Control into the corresponding labeled test tube.

Use a new tip for each calibrator and control. The same tip can be used if running a duplicate

Must remove all air bubbles from tip before drawing final sample

Step 3: Adding Reagents & Vortexing

- Before dispensing reagents, gently swirl the contents of each reagent bottle.
- To eliminate air bubbles and oxidized reagent, dispense 5-6 aliquots of each reagent into a waste container immediately before use.
- Dispense 1 aliquot of FaSafe™ Reagent A into every calibrator and control test tube.
- Dispense 1 aliquot of FaSafe™ Reagent B into every calibrator and control test tube.
- Once you have aliquoted Reagent B into the last test tube start the timer for 5 minutes.
- Cap the test tubes and vortex them at the fastest dial setting for 15 seconds.
- Place the test tubes in the heat block at 40°C for the remaining time.

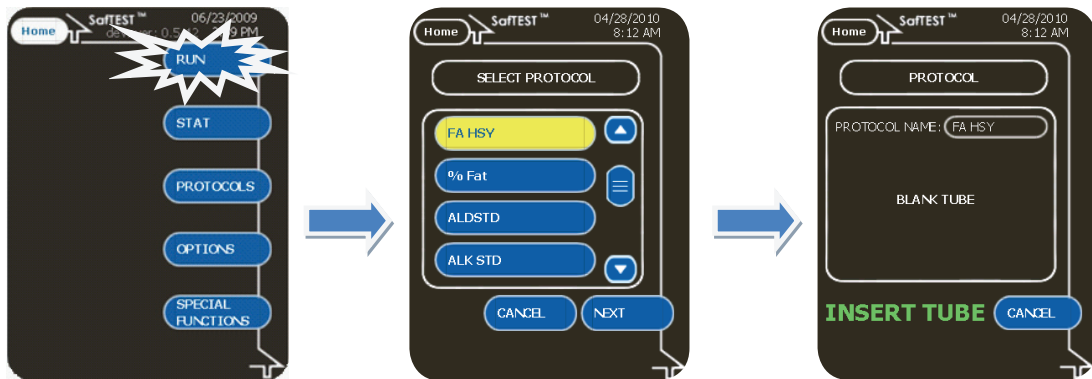
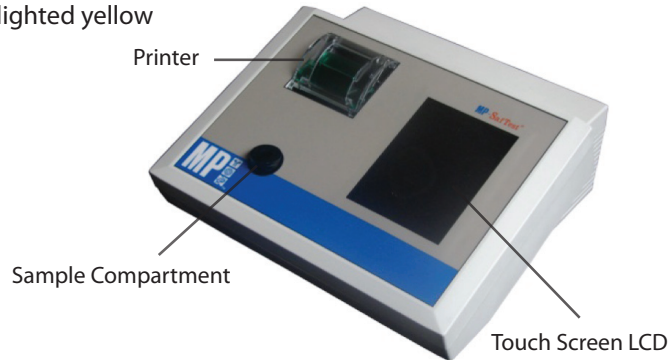
***Once time has ended, IMMEDIATELY invert sample and read in SafTest analyzer.
If samples are not read immediately then the tubes must be discarded and test should be rerun***

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Step 4: SafTest Analyzer Set Up with Calibration [RUN]

- Press [RUN] icon on the Touch Screen LCD.
- Press [FAHSY] icon, when highlighted yellow press [NEXT] icon.



Immediately before inserting each test tube in the analyzer, wipe the outside of the tube with a Kimwipe. Fingerprints or debris on the tube may result in an inaccurate reading

- At the "Blank Tube" prompt, insert a test tube filled half way with distilled water in the sample compartment. Do not remove tube until prompted or after beep.
- At the "Cal #_ of 4" prompts, insert Calibrators 1-4 in the sample compartment.

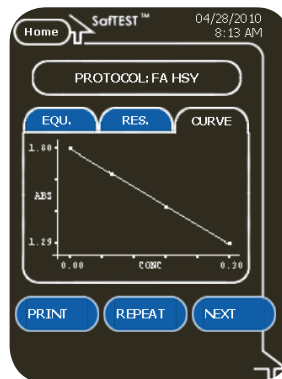


Figure 1. FAHSY calibration screen shot.

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- At the “DO YOU WANT TO STORE THE CALIBRATORS” prompt, select “YES” (Figure 2).
- Select the number of replicates to be analyzed (1-5). Use the “+” or “-” arrows to select a number (normally duplicates, i.e. 2, should be analyzed to ensure results are accurate and consistent). Press the [NEXT] icon.
- At the “Smpl #_ Rep #_, Insert Tube” prompt, insert the duplicate control tubes into the sample compartment.
- SafTest AnalyzerII will continue asking for samples until the user quits the program by pressing [STOP] and then [DONE].

*****The SafTest AnalyzerII arbitrarily numbers each sample 1-99.
Be sure to re-label what sample you are running*****

- Verify that the control fall within the FaSafe HSY control range which can be found on the “FaSafe HSY Control Package Insert”. Your test values for the controls should approximate these ranges. If they do not, rerun the test.

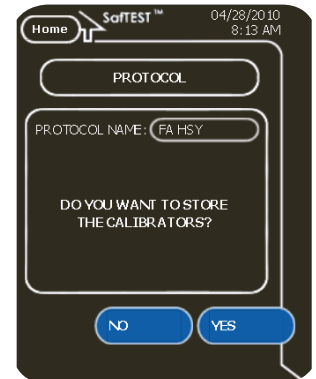


Figure 2. FASTD store calibrators screen shot.

Step 5: Analyzing Samples

- Samples should be prepared according to “Preparing Samples for STD Assays”.
- Label two new 12mm glass test tubes per sample: S1 and S2 (for running duplicates).
- Use a positive displacement pipette to transfer 100µl of each prepared sample into the corresponding labeled test tube.
- Dispense 1 aliquot of FaSafe™ Reagent A into every sample test tube.
- Dispense 1 aliquot of FaSafe™ Reagent B into every sample test tube.
- Once you have aliquoted Reagent B into the last test tube start the timer for 5 minutes.
- Cap the test tubes and vortex them at the fastest dial setting for 15 seconds.
- Place the test tubes in the heat block at 40°C for the remaining time.
- Invert tubes before reading them on the Analyzer.

Step 6: SafTest Analyzer Set Up without Calibration [STAT]

- Press [STAT] icon.
- Press [FAHSY] icon, when highlighted yellow press [NEXT] icon.
- At the “Blank Tube” prompt, insert a test tube of distilled water in the sample compartment.
- Select the number of replicates to be analyzed. Use the “+” or “-” arrows to select a number (Normally two replicates should be analyzed to ensure that results are consistent). Press [NEXT] icon.
- At the “Smpl #_ Rep #_, Insert Tube” prompt, insert control or sample tubes in the sample compartment.
- SafTest Analyzer will continue asking for samples until the user quits the program by pressing [STOP] and then [DONE].

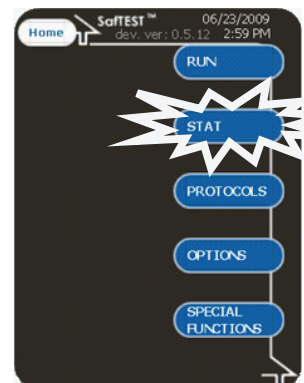


Figure 3. Home screen shot.

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Step 7: Calculating Your Results

- The SafTest Analyzer will use the calibrators to calculate the acid content as percent (%) oleic acid in the sample.
- Adjust instrument results by taking into account the dilution factor. For example:

Dilution Factor	SafTest Results	Dilution x Results	Final Results
1:4	0.19%	4 x 0.19%	0.76%

- The final result can be calculated to % oleic acid on a fat basis by taking the percent fat into consideration when performing your calculation. For example, with a sample that is 15% fat and yields a final result of 0.76%:

Final Result	Result/(% Fat/100)	Final Result
0.76%	0.76/(15/100)	5.07%

Troubleshooting

- If the sample value is greater than the value of the highest calibrator, the instrument will flag the results as “HI.” The sample must be diluted at a higher dilution and retested; see **Preparing Additional Dilutions Card**. Values that are flagged “HI” are inaccurate and should be rerun at a higher dilution.
- If the sample value is less than the value of the lowest calibrator, the instrument will flag the results as “LO”. Values that are flagged “LO” should be reported as “< (value of the lowest calibrator).”
- Check the instrument printout for flags or error messages before reporting results. The coefficient of variation (%CV) should be less than 10%. Higher variations will be flagged, in which case you should repeat the test.
 - EXCEPTION** – A large %CV is expected with samples that are measured at the low end of the calibration curve. For such measurements, do not repeat the test.
- Ranges for the control are found on the “FaSafe HSY Control Package Insert,” which come with each kit. Your test values for the controls should approximate these ranges. If they do not, rerun the test.
- If a STAT curve is going to be used, make sure that at least one control is run at the beginning of each day of testing. It is recommended that a new calibration curve be stored weekly.

Step 8: Storing Equipment

- At the end of the day, store the calibrators, controls, and reagent bottles (with dispensers attached) at 2-6°C.
- To maximize lamp life, turn OFF the SafTestII Analyzer when not in use. Power OFF the SafTest Analyzer by pressing the [O] marked portion of the power switch, which is located on the rear panel of the SafTestII Analyzer.