Triglyceride Assay; Single Cuvette (Pointe Scientific)

Materials Required:

1. Pipettes: 10uL – 1000uL
2. Test tubes and racks
3. Timer
4. Water bath (37°C)
5. Triglyceride GPO Reagent (Pointe Scientific Cat #T7532)
6. Triglyceride Standard (Pointe Scientific Cat #T7531)
7. Quality Controls (Multi-analyte Cholestech #88773)
8. Cary Spectrophotometer set at 500nm (see 'Cary Spectrophotometer_Single Cuvette' instructions)
9. Cuvettes

Sample Collection/ Storage:

1. Clear, unhemolyzed serum is sample of choice.
2. Serum (gold-topped tube): Gently invert vial 8-10x immediately following collection, let sit 30min at RT then centrifuge at 1000g's (2.6-3.0 RPM) for 15min.
3. Following centrifugation, immediately pipette serum into appropriately labeled micro-centrifuge tubes.
4. Samples should be run immediately or frozen for batch analyses.

Procedure:

1. Turn on water bath to 37°C.
2. Label test tubes, in duplicate, and place in rack (e.g. Blank, Standard, Control, Subject ID).
3. Pipette 1000uL of GPO reagent into each tube and place in water bath for 5 minutes. The 'blank' tube will only receive the reagent.
4. Add 10uL of standard/control/sample into remaining tubes and vortex using low setting.
5. Place tubes back in water bath and incubate for 5 minutes.
6. During incubation prepare the spectrophotometer. (Refer to 'Cary Spectrophotometer_Single Cuvette' instructions).
7. Zero spectrophotometer using the blank at 500nm.
8. Read the absorbance of all the tubes and enter in excel template (Triglyceride Assay_Single Cuvette_Results Template).

Notes: Final color is stable for 60 minutes.

Reportable Range (linearity) is 0 – 1000mg/dL. If a sample exceeds the upper linearity limit it needs to be diluted with isotonic saline and rerun. Multiply results by 2.

Verifying Results:

1. Make sure your controls are within range.
2. Verify that your coefficient of variation for your sample duplicates are within acceptable limits ($\leq$15%), rerun if necessary.