**LCAT Activity Assay Protocol**

1. In a 1.5 mL centrifuge tube, mix HDL and LCAT Assay buffer reagent according to excel sheet.
2. Add HDL + LCAT Assay buffer into well of polypropylene reaction plate. (Total Volume 50 uL)
3. Add 50 uL Substrate + LCAT Assay buffer into each well.
   1. 46 subjects, 1 control, 1 blank = 47 x2 = 96 wells
      1. 96+9 = 105
      2. **MIX**
         1. **Substrate**: 0.5 ul substrate x 105 = **52.5 ul**
      3. **Buffer**: 5250 ul – 52.5 = **5197.5 ul**
4. Incubate for 2.5 HR in 37C microplate incubator.
5. Add 200 uL of Read reagent to each well.
   1. 200 uL \* 105 = **21000 uL read reagent** in a reservoir
6. Transfer 200 uL of the reaction mixture from the polypropylene plate to a black fluorescence microplate
7. Measure fluorescence at ex = 340/em = 390 and 470 nm