



UC Davis MMPC-Live Protocol

Hepatic Triglyceride Production

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Replaces version: None

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Summary:

Triglycerides are water-insoluble energy-rich lipids secreted by the liver as part of very low density lipoproteins (VLDLs) to supply energy to a variety of extrahepatic tissues. In 1951, Aaron Kellner and colleagues described IV injection of nonionic detergents resulted in a "sustained hyperlipemia". It was later shown that this was due to the inhibition of TG hydrolysis by lipoprotein lipase (LPL). Lipolysis inhibition has since been used regularly to determine hepatic TG production rates, with both Kolliphor P 407 micro and Triton WR-1339 being widely used.

Reagents and Materials:

<i>Reagent/Material</i>	<i>Vendor</i>	<i>Stock Number</i>
Microvette CB 300 Lithium heparin LH blood collection tube	Sarstedt	16.443
Microcentrifuge tubes		
Sterile normal saline		

Protocol:

1. If desired for the study design, fast mice into clean cages starting at lights on.

Fasting lowers baseline TG levels slightly but typically does not affect the rate of triglyceride (TG) production.

2. Collect a 50 μ l baseline blood sample from each mouse. Flick tube to mix and place on ice.
3. Inject mice with either Kolliphor P 407 micro (1,000 mg/kg IP; Sigma) or Triton WR-1339 (500 mg/kg IV; Sigma) in saline approximately 6 h into the light cycle.
4. Collect 50 μ l blood samples from each mouse at 1, 2, and 4 h following injection. Flick tube to mix and place on ice.
5. Prepare plasma by centrifuging at 2,000 x g for 10 minutes and transferring supernatant to a fresh labeled microfuge tube.
6. Store samples at $< -20^{\circ}\text{C}$ until triglyceride (TG) levels are measured.
7. TG levels are measured using the UCD_MMHcore_Triglycerides protocol.
8. TG production rate is calculated as the difference in plasma TG levels between given intervals following detergent injection.

Reagent Preparation:

Dissolve either P-407 or Triton detergent in saline at 10 mg/ml. Filter sterilize.