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Protocol status: Working
 We use this protocol and it's working

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 21344

Yale - Non-Esterified Fatty Acids

John Stack¹, Gary Cline¹

¹Yale University

Mouse Metabolic Phenotyping Centers
 Tech. support email: info@mmpc.org

Lili Liang

ABSTRACT

Summary:

Procedure used to determine the concentration of NEFA in blood, serum, and plasma. NEFA; Free fatty acids are measured in a multistep reaction to form an colored adduct of 3-methyl-Nethyl-N-(b-hydroxy-ethyl)-aniline and 4-aminoantipyrine monitored at 560 nm.

MATERIALS

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⊗ NEFA Reagents A & B **Wako Catalog #H7587-58**

⊗ NEFA Solvents A & B **Wako Catalog #H7587-58**

Reagent Preparation:

NEFA Reagent A: Add 50 mL of Solvent A to Reagent A. Gently invert and allow 15 minutes to mix.

NEFA Reagent B: Add 25 mL of Solvent B to Reagent B. Gently invert and allow 15 minutes to mix.

NEFA Solvents A & B: As supplied by vendor.

Note:

Wako [RRID:SCR_013651](#)

BEFORE START INSTRUCTIONS

Analysis by automated system Cobas Mira Plus

Keywords: Non-Esterified
Fatty Acids (NEFA)

- 1 Calibrate Cobas for NEFA analysis by running a NEFA standard.

- 2 Sample handling as performed by the Cobas Mira Plus
 - a) Pipette 6µL of sample into cuvette.
 - b) Add 225 µL of NEFA Reagent A Mixture.
 - c) Add 75 µL of NEFA Reagent B Mixture.
 - d) Mixture is incubated at 37°C for 10 minutes
 - e) Absorbance is measured at 560 nm.