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Yale - Alkaline Phosphatase

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

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Abstract

Summary:

Procedure to measure the amount of Alkaline Phosphatase activity. Alkaline Phosphatase (ALP) activity is measured from the hydrolysis of 4-nitrophenylphospate to 4-nitrophenyoxide ion (monitored at 405 nm) and phosphate.

Materials

MATERIALS

Alkaline Phosphatase Reagent **Prolabs(cliniqa)** **Catalog #**R85120

Assayed Control Serum 1 **Prolabs(cliniqa)** **Catalog #**R83082

Assayed Control Serum 2 **Prolabs(cliniqa)** **Catalog #**R83083

Reagent Preparation:

Alkaline Phosphatase Reagent: Add the appropriate amount of water (6.5mL) to the reagent bottle. Invert to mix, allowing 15 minutes for the reagent to settle.

Assayed Control Serum 1: Add the appropriate amount of water (6.5mL) to the chemical control bottle. Invert to mix, allowing 15 minutes for the reagent to settle.

Assayed Control Serum 2: Add the appropriate amount of water (6.5mL) to the chemical control bottle. Invert to mix, allowing 15 minutes for the reagent to settle.

Before start

Analysis by automated system Cobas Mira Plus.

1 Calibrate Cobas for Alkaline Phosphatase Activity analysis by running two assayed control serum.

2 Sample handling as performed by the Cobas Mira Plus.
a) Pipette 3 µL of sample into a cuvette slot.
b) Add 150 µL of Alkaline Phosphatase Reagent.
c) Mixture is incubated at 37˚C and spun for 10 minutes.
d) Absorbance is measured at 405 nm.